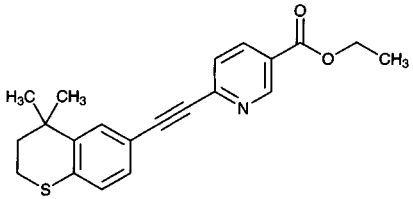
Tazarotene



Molecular formula: $C_{21}H_{21}NO_2S$
Molecular weight: 351.47
CAS Registry No.: 118292-40-3
Merck Index: 9249

SAMPLE

Matrix: blood, microsomal incubations
Sample preparation: Blood. 500 µL Whole blood + 2.5 mL MeCN:1-butanol 50:50 + 200 µL 1 µg/mL IS, vortex for 30 s, centrifuge at 1500 g for 5 min. Evaporate supernatant to dryness with nitrogen. Microsomal incubations. 1 mL Microsomal incubation + 2.5 mL MeCN:1-butanol 50:50, centrifuge at 1500 g for 5 min. Decant the supernatant, dry it with nitrogen and reconstitute in 300 µL mobile phase. Inject a 100 µL aliquot.

HPLC VARIABLES

Guard column: dry packed C18
Column: 150 × 4.6 5 µm Ultrasphere C18
Mobile phase: MeCN:water:acetic acid 65:34.5:0.5
Flow rate: 1.2
Injection volume: 100
Detector: UV 345

CHROMATOGRAM

Retention time: 17
Internal standard: AGN 190252 (7)

OTHER SUBSTANCES

Extracted: metabolites

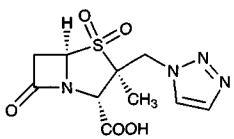
KEY WORDS

liver; human; rat; whole blood

REFERENCE

Madhu,C.; Duff,S.; Baumgarten,V.; Rix,P.; Small.D.,; Tang-Liu,D. Metabolic deesterification of tazarotene in human blood and rat and human liver microsomes, *J.Pharm.Sci.*, **1997**, *86*, 972–974.

Tazobactam



Molecular formula: $C_{10}H_{12}N_4O_5S$
Molecular weight: 300.30
CAS Registry No.: 89786-04-9
Merck Index: 9251
Lednicer No.: 5 156

SAMPLE

Matrix: bile, blood, urine
Sample preparation: Plasma, serum. 200 µL Plasma or serum + 200 µL 25 µg/mL penicillin G in 50 mM pH 6.0 sodium phosphate buffer + 800 µL MeCN, vortex for 30 s, centrifuge at 3000 g for 10 min. Remove the clear supernatant and add it to 2 mL dichloromethane, mix for 30 s, centrifuge at 3000 g for 10 min, inject a 25 µL aliquot of the upper aqueous layer. Bile. 200 µL Bile + 400 µL 50 mM pH 7.0 sodium phosphate buffer + 2 mL MeCN, vortex for 30 s, centrifuge at 3000 g for 10 min. Remove the clear supernatant and add it to 2 mL dichloromethane, mix for 30 s, centrifuge at 3000 g for 10 min, inject a 25 µL aliquot of the upper

aqueous layer. Urine. 100 μL Urine + 50 μL 5 mg/mL penicillin G in water, vortex for 30 s, make up to 10 mL with 50 mM pH 6.0 sodium phosphate buffer, inject a 25 μL aliquot.

HPLC VARIABLES
Guard column: 15 × 3.2 7 μm Brownlee C 18 guard column
Column: 250 × 4.6 5 μm Hypersil ODS (Keystone)
Mobile phase: Gradient. A was MeCN:10 mM sodium phosphate adjusted to pH 2.7 with concentrated phosphoric acid 3:97. B was MeCN:10 mM sodium phosphate adjusted to pH 2.7 with concentrated phosphoric acid 90:10. A:B from 95:5 to 50:50 over 9 min and then to 95:5 over 1 min.
Flow rate: 1.5
Injection volume: 25
Detector: UV 220

CHROMATOGRAM
Retention time: 5.7
Internal standard: penicillin G (12.5)
Limit of quantitation: 50000 ng/mL (urine), 1000 ng/mL (plasma, serum, bile)

OTHER SUBSTANCES
Extracted: piperacillin
Simultaneous: amoxicillin, ampicillin, cefoperazine, cefometazole, cefotaxime, cefotetan, cefuroxime, mezlocillin

KEY WORDS
plasma; serum

REFERENCE
Ocampo,A.P.; Hoyt,K.D.; Wadgaonkar,N.; Carver,A.H.; Puglisi,C.V. Determination of tazobactam and piperacillin in human plasma, serum, bile and urine by gradient elution reversed-phase high-performance liquid chromatography, *J.Chromatogr.*, **1989**, *496*, 167–179.

SAMPLE
Matrix: blood
Sample preparation: Homogenize tissue in water at 4° for 30 s, centrifuge at 15000 rpm (Sorvall centrifuge) for 10 min. 250 μL Plasma or tissue homogenate + 500 μL 10 μg/mL cefpodoxime in MeCN, mix, centrifuge at 15000 rpm (Sorvall centrifuge), extract with 1 mL dichloromethane, inject 120 μL of the aqueous phase onto column A with mobile phase A, elute the contents of column A onto column B with mobile phase B, monitor the effluent from column B.

HPLC VARIABLES
Column: A 40 × 4.6 10 μm Lichrosorb RP 2; B 250 × 4.6 5 μm Spherisorb ODS II
Mobile phase: A MeCN:100 mM NaH_2PO_4 and 5 mM tetrabutylammonium hydrogen sulfate 5:95, pH adjusted to 6.5; B MeCN:100 mM NaH_2PO_4 and 5 mM tetrabutylammonium hydrogen sulfate 10:90, pH adjusted to 6.5
Column temperature: 25
Flow rate: A 1; B 1.5
Injection volume: 120
Detector: UV 210 (UV 300 for IS)

CHROMATOGRAM
Retention time: 18.6
Internal standard: cefpodoxime (24.9)
Limit of detection: 96 ng/mL

KEY WORDS
plasma; column-switching

REFERENCE
Kinzig,M.; Sörgel,F.; Brismar,B.; Nord,C.E. Pharmacokinetics and tissue penetration of tazobactam and piperacillin in patients undergoing colorectal surgery, *Antimicrob.Agents Chemother.*, **1992**, *36*, 1997–2004.

SAMPLE
Matrix: blood
Sample preparation: 1 mL Serum + 400 μL 10 mg/mL zinc sulfate containing 350 μg/mL benzoic acid, vortex for 30 s, centrifuge at 5500 g for 5 min, inject a 20 μL aliquot of the supernatant.

HPLC VARIABLES
Column: 150 × 4.3 5 μm Nova Pak
Mobile phase: MeOH:pH 6.30 phosphate buffer 5:95
Column temperature: 45
Flow rate: 2
Injection volume: 20
Detector: UV 225

CHROMATOGRAM
Retention time: 2.4
Internal standard: benzoic acid (1.9)
Limit of detection: 5 μg/mL

OTHER SUBSTANCES
Extracted: sulbactam

KEY WORDS
serum

REFERENCE
Guillaume,Y.; Peyrin,E.; Guinchard,C. Rapid determination of sulbactam and tazobactam in human serum by high-performance liquid chromatography, *J.Chromatogr.B*, **1995**, *665*, 363–371.

SAMPLE
Matrix: blood
Sample preparation: 1 mL Serum or hemofiltrate + 100 μL 3 M sulfuric acid + 4 mL diethyl ether, vortex for 1 min, centrifuge at 5500 g for 10 min. Remove the organic layer and evaporate it to dryness under a stream of nitrogen at 37°, reconstitute the residue in 100 μL reagent and 10 μL 1% mercuric chloride in water, heat at 37° for 30 min, inject a 20 μL aliquot. (Prepare reagent by dissolving 4 g 1,2,4-triazole in water, adjusting the pH to 9.0 with 10 M NaOH, and making up to 20 mL with water.)

HPLC VARIABLES
Column: 125 × 4 5 μm Lichrospher RP (18)
Mobile phase: MeOH:$(NH_4)_2HPO_4$ buffer 0.5:99.5, adjusted to pH 6.00 with phosphoric acid (Buffer concentration not given.)
Flow rate: 2.2
Injection volume: 20
Detector: UV 325

CHROMATOGRAM
Retention time: 8
Limit of detection: 50 ng/mL

KEY WORDS
serum; hemofiltrate; derivatization

REFERENCE
Peyrin,E.; Guillaume,Y.; Guinchard,C. High-performance liquid chromatographic determination of tazobactam by precolumn derivatization, *J.Chromatogr.B*, **1995**, *672*, 160–164.

SAMPLE
Matrix: blood, tissue

Sample preparation: Plasma. 250 μL Plasma + 500 μL 10 μg/mL cefpodoxime in MeCN, mix, centrifuge at 15000 rpm (Sorvall centrifuge). Remove the supernatant and add it to 1 mL dichloromethane, extract, inject a 120 μL aliquot of the aqueous phase. Tissue. Blot tissue, homogenize with two volumes of water (IKA-Ultra-Turrax) at 4° for 30 s, centrifuge at 15000 rpm (Sorvall centrifuge) for 10 min, remove 250 μL of the supernatant, add 500 μL 10 μg/mL cefpodoxime in MeCN, mix, centrifuge at 15000 rpm (Sorvall centrifuge). Remove the supernatant and add it to 1 mL dichloromethane, extract, inject a 120 μL aliquot of the aqueous phase. (A column-switching technique is used but no details are given in the paper.)

HPLC VARIABLES
Column: A 40 × 4.6 10 μm RP-2 (Bischoff Chromatography)); B 250 × 4.6 5 μm Spherisorb ODS II
Mobile phase: A MeCN:100 mM NaH_2PO_4 + 5 mM tetrabutylammonium hydrogen sulfate 5:95, pH 6.5; B MeCN:100 mM NaH_2PO_4 + 5 mM tetrabutylammonium hydrogen sulfate 10:90, pH 6.5
Column temperature: 25
Flow rate: A 1; B 1.5
Injection volume: 120
Detector: UV 210 for tazobactam, UV 300 for cefpodoxime

CHROMATOGRAM
Retention time: 18.6
Internal standard: cefpodoxime (24.9)
Limit of quantitation: 76 ng/g (tissue), 96 ng/mL (plasma)

KEY WORDS
plasma; column-switching; pharmacokinetics; fat; muscle; skin; intestinal mucosa; appendix

REFERENCE
Kinzig,M.; Sörgel,F.; Brismar,B.; Nord,C.E. Pharmacokinetics and tissue penetration of tazobactam and piperacillin in patients undergoing colorectal surgery, *Antimicrob.Agents Chemother.*, **1992**, *36*, 1997–2004.

SAMPLE
Matrix: formulations
Sample preparation: Dilute 18:1 with mobile phase, inject a 20 μL aliquot.

HPLC VARIABLES
Column: 250 × 4.6 5 μm Adsorbosphere
Mobile phase: MeCN:10 mM NaH_2PO_4 7:93, pH adjusted to 2.7 with 85% phosphoric acid
Flow rate: 1.2
Injection volume: 20
Detector: UV 215

CHROMATOGRAM
Retention time: 5.0

KEY WORDS
injections; saline; stability-indicating

REFERENCE
Choi,J.-S.; Burm,J.-P.; Jhee,S.S.; Chin,A.; Ulrich,R.W.; Gill,M.A. Stability of piperacillin sodium-tazobactam sodium and ranitidine hydrochloride in 0.9% sodium chloride injection during simulated Y-site administration, *Am.J.Hosp.Pharm.*, **1994**, *51*, 2273–2276.

SAMPLE
Matrix: formulations
Sample preparation: Dilute 8-fold to 20-fold with saline, filter (0.2 μm), inject a 20 μL aliquot of the filtrate.

HPLC VARIABLES
Column: 250 × 4.6 5 μm Adsorbosphere C18

Mobile phase: MeCN:100 mM sodium phosphate buffer 7:93 adjusted to pH 2.7 with 85% phosphoric acid
Flow rate: 1.2
Injection volume: 20
Detector: UV 215

CHROMATOGRAM
Retention time: 7-8

KEY WORDS
saline; injections; stability-indicating

REFERENCE
Chung,K.C.; Moon,Y.S.K.; Chin,A.; Ulrich,R.W.; Gill,M.A. Compatibility of ondansetron hydrochloride and piperacillin sodium tazobactam sodium during simulated Y-site administration, *Am.J.Health-Syst.Pharm.*, **1995**, *52*, 1554–1556.

SAMPLE
Matrix: formulations
Sample preparation: Dilute a 25-50 μL sample with 2 mL mobile phase, filter (0.2 μm), inject a 20 μL aliquot.

HPLC VARIABLES
Column: 250 × 4.6 5 μm Adsorbosphere C18
Mobile phase: MeCN:10 mM sodium phosphate 7:93 adjusted to pH 2.7 with 85% phosphoric acid
Flow rate: 1.2
Injection volume: 20
Detector: UV 215

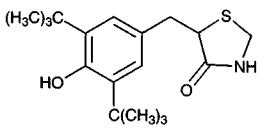
CHROMATOGRAM
Retention time: 6.40

OTHER SUBSTANCES
Simultaneous: degradation products

KEY WORDS
injections; saline; 5% dextrose

REFERENCE
Moon,Y.S.K.; Chung,K.C.; Chin,A.; Gill,M.A. Stability of piperacillin sodium-tazobactam sodium in polypropylene syringes and polyvinyl chloride minibags, *Am.J.Health-Syst.Pharm.*, **1995**, *52*, 999–1001.

Tazofelone



Molecular formula: $C_{18}H_{27}NO_2S$
Molecular weight: 321.49
CAS Registry No.: 136433-51-7

SAMPLE
Matrix: microsomal incubations
Sample preparation: Add 1 mL cold MeCN to 250 μL microsomal incubation, centrifuge at 13000 rpm for 3 min, evaporate the supernatant under nitrogen, reconstitute the residue in 100 μL MeCN:water 40:60, inject 60 μL aliquot.

HPLC VARIABLES
Column: 250 × 4.6 Prodigy C 18 (Phenomenex)

Mobile phase: MeCN:water 60:40
Column temperature: 40
Flow rate: 1
Injection volume: 60
Detector: UV 214

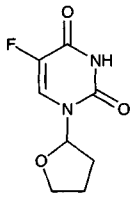
CHROMATOGRAM
Retention time: 15

OTHER SUBSTANCES
Extracted: metabolites

KEY WORDS
human; liver

REFERENCE
Surapaneni,S.S.; Clay,M.P.; Spangle,L.A.; Paschal,J.W.; Lindstrom,T.D. In vitro biotransformation and identification of human cytochrome P450 isozyme-dependent metabolism of tazofelone, *Drug Metab.Dispos.*, **1997**, *25*, 1383–1388.

Tegafur



Molecular formula: $C_8H_9FN_2O_3$
Molecular weight: 200.17
CAS Registry No.: 17902-23-7
Merck Index: 9267

SAMPLE
Matrix: blood
Sample preparation: Mix 20 μL plasma with 50 μL 500 mM pH 8.0 phosphate buffer, 100 μL 500 ng/mL 5-chlorouracil in water, and 7 mL ethyl acetate. Shake for 30 min and centrifuge at 2200 g for 10 min. Remove the ethyl acetate layer and evaporate it. Reconstitute the residue in 500 μL n-hexane:mobile phase 40:60 with sonication for 10 min. Inject the whole amount.

HPLC VARIABLES
Column: 100 × 4.6 Develosil 60-3 (Nomura Chemical, Japan)
Mobile phase: n-Hexane:ethyl acetate:formic acid:water 50:0.5:0.5:0.3
Flow rate: 0.9
Injection volume: 500
Detector: UV 264

CHROMATOGRAM
Internal standard: 5-chlorouracil
Limit of detection: 50 ng/mL

KEY WORDS
plasma; rat; normal phase

REFERENCE
Fuse,E.; Takai,K.; Okuno,K.; Kobayashi,S. Hepatic extraction ratio of 5-fluorouracil in rats. Dose dependence and effect of uracil and interleukin-2, *Biochem.Pharmacol.*, **1996**, *52*, 561–568.

SAMPLE
Matrix: blood, urine
Sample preparation: Dilute 250 μL plasma or urine 1:10 with water, add 100 μL 200 mM pH 7.0 phosphate buffer, add 100 μL 10 μg/mL β-hydroxyethyltheophylline, add 4 mL dichloromethane, shake for 10 min, centrifuge at 2000 g for 5 min, transfer the organic layer to another

tube, repeat the extraction. Evaporate the combined organic layers to dryness under a stream of nitrogen at 40°, dissolve the residue in 200 μL mobile phase, inject a 40 μL aliquot.

HPLC VARIABLES
Column: 150 × 4.6 5 μm Inertsil ODS-2 (GL Sciences)
Mobile phase: MeOH:10 mM pH 5.5 phosphate buffer 15:85
Flow rate: 1
Injection volume: 40
Detector: UV 270

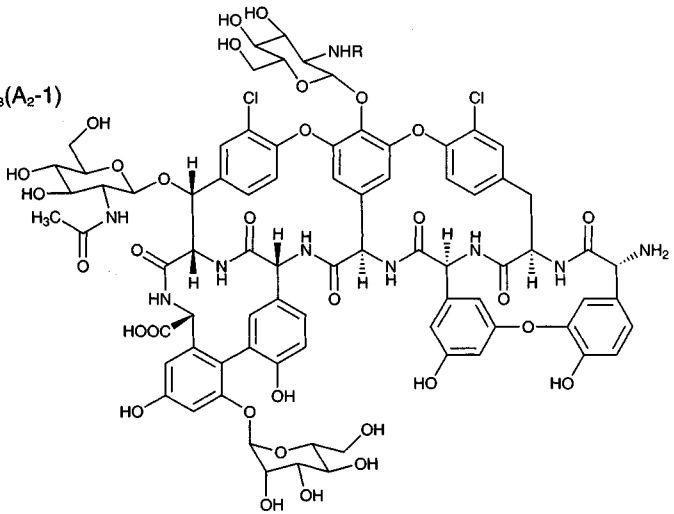
CHROMATOGRAM
Retention time: 6.95
Internal standard: β-hydroxyethyltheophylline (10.53)
Limit of quantitation: 10 ng/mL (plasma), 100 ng/mL (urine)

KEY WORDS
plasma

REFERENCE
Matsushima,E.; Yoshida,K.; Kitamura,R.; Yoshida,K.-. Determination of S-1 (combined drug of tegafur, 5-chloro-2,4-dihydroxypyridine and potassium oxonate) and 5-fluorouracil in human plasma and urine using high-performance liquid chromatography and gas-chromatography-negative ion chemical ionization mass spectrometry, *J.Chromatogr.B*, **1997**, *691*, 95–104.

Teicoplanin

Molecular formula: $C_{88}H_{95}Cl_2N_9O_{33}$($A_2$-1)
Molecular weight: 1877.68 (A_2-1)
CAS Registry No.: 61036-64-4, 61036-62-2
Merck Index: 9269



Teicoplanin A_2-1 (Z)-4-decanoic acid
A_2-2 8-methylnonanoic acid
A_2-3 n-decanoic acid
A_2-4 8-methyldecanoic acid
A_2-5 9-methyldecanoic acid

SAMPLE
Matrix: blood
Sample preparation: Add 1 mL MeCN to 500 μL plasma, vortex briefly, centrifuge at 2000 g for 3 min, add 2 mL chloroform (Caution! Chloroform is a carcinogen!) to the supernatant, vortex, inject an aliquot of the aqueous supernatant layer.

HPLC VARIABLES
Column: 250 × 4 5 μm LiChrocart RP8

Mobile phase: MeCN:20 mM pH 4.4 ammonium acetate buffer 26:74
Flow rate: 1.3
Injection volume: 30
Detector: UV 220

CHROMATOGRAM
Retention time: 6.5
Limit of detection: 30 ng/mL
Limit of quantitation: 90 ng/mL

KEY WORDS
plasma

REFERENCE
Cociglio,M.; Peyrière,H.; Hillaire-Buys,D.; Alric,R. Application of a standardized coextractive cleanup procedure to routine high-performance liquid chromatography assays of teicoplanin and ganciclovir in plasma, *J.Chromatogr.B*, **1998**, *705*, 79–85.

SAMPLE
Matrix: blood
Sample preparation: Condition a Bond Elut SAX SPE cartridge with 4 mL MeOH, 1 mL water, and 1 mL 10 mM n-heptanesulfonic acid. 1 mL Plasma + 1 mL 10 mM n-heptanesulfonic acid, vortex, centrifuge at 4000 g for 2 min, add the supernatant to the SPE cartridge, wash with 3 mL 10 mM heptanesulfonic acid, elute with 1 mL MeOH, inject a 20 μL aliquot of the eluate.

HPLC VARIABLES
Guard column: 10 × 4.2 10 μm LiChrosorb RP-8
Column: 150 × 4.2 5 μm LiChrosorb RP-8
Mobile phase: MeOH:water 5:95 containing 10 mM disodium n-heptanesulfonate, adjusted to pH 4.0 with 2 g/L sodium acetate and glacial acetic acid
Column temperature: 30
Flow rate: 0.8
Injection volume: 20
Detector: UV 240

CHROMATOGRAM
Retention time: 6.5
Limit of detection: 200-400 ng/mL

KEY WORDS
SPE; plasma

REFERENCE
Georgopoulos,A.; Czejka,M.J.; Starzengruber,N.; Jäger,W.; Lackner,H. High-performance liquid chromatographic determination of teicoplanin in plasma: comparison with a microbiological assay, *J.Chromatogr.*, **1989**, *494*, 340–346.

SAMPLE
Matrix: blood
Sample preparation: Add plasma to a Bond Elut C8 SPE cartridge, wash with water, elute with MeCN:pH 6 buffer 20:80, inject a 50 μL aliquot of the eluate into a 500 μL sample lop filled with water, inject 200 μL water into the sample loop, operate the valve.

HPLC VARIABLES
Column: 100 × 1 5 μm Hypersil ODS
Mobile phase: MeCN:25 mM pH 6.0 NaH_2PO_4 20:80
Flow rate: 0.05
Injection volume: 50
Detector: UV 210

CHROMATOGRAM
Retention time: 3 (TA3-1), 10 (TA2-1), 13 (TA2-2), 16 (TA2-3), 30 (TA2-4), 33 (TA2-5)

Limit of detection: 50 ng/mL

KEY WORDS
SPE; plasma

REFERENCE
Taylor,R.B.; Reid,R.G.; Gould,I.M. Determination of teicoplanin in plasma using microbore high-performance liquid chromatography and injection-generated gradients, *J.Chromatogr.*, **1991**, *563*, 451–457.

SAMPLE
Matrix: blood, urine
Sample preparation: Add 1-10 mL plasma or urine to the affinity column, wash with 1 mL buffer, elute with three 500 μL portions of 1.5% ammonium hydroxide. Add 100 μL 18% HCl to the eluate, inject a 170 μL aliquot. (Affinity columns consisted of 0.5 mL D-alanyl-D-alanine-ε-aminocaproyl Sepharose CL-6B stored in 100 mM pH 8.5 Tris buffer containing 0.004% sodium merthiolate, equilibrate with buffer before use. After use wash with 5 mL buffer containing 0.004% sodium merthiolate, store at 4°. Buffer was 50 mM NaH_2PO_4 containing 200 mM NaCl adjusted to pH 7.5 with 1 M NaOH. Prepare the column material by coupling D-alanyl-D-alanine to activated CH-Sepharose 4B using ε-aminocaproic acid according to the manufacturer's instructions. 30 mg D-Alanyl-D-alanine in 5 mL 100 mM sodium bicarbonate buffer containing 500 mM NaCl (pH 8) was coupled to 3 mL gel in 1 h, block unreacted ester groups with 1 M ethanolamine hydrochloride at pH 9 for 1 h, wash repeatedly with 100 mM pH 4 sodium acetate buffer containing 500 mM NaCl and 100 mM pH 8 tris-HCl buffer containing 500 mM NaCl (Appl. Biochem. Biotechnol. 1985, 11, 101.).)

HPLC VARIABLES
Guard column: 30 × 4.6 5 μm Nucleosil C18
Column: 100 × 4.6 5 μm Nucleosil C18
Mobile phase: Gradient. A was 10 mM pH 4.9 NaH_2PO_4. B was MeCN:10 mM pH 4.9 NaH_2PO_4 50:50. A:B 78:22 of 0.5 min, to 47:53 over 44 min, maintain at 47:53 for 2 min, to 0:100 over 1.5 min, maintain at 0:100 for 5 min, return to initial conditions over 2 min, re-equilibrate for 5 min.
Flow rate: 1.3
Injection volume: 170
Detector: UV 240

CHROMATOGRAM
Retention time: 9.1 (A3), 28.8 (A2-1), 31.6 (A2-2), 32.8 (A2-3), 37.1 (A2-4), 38.0 (A2-5)
Limit of detection: 100 ng/mL (A2-2)

OTHER SUBSTANCES
Noninterfering: acebutolol, acetaminophen, aspirin, atenolol, benfluorex, chlorazepam, clofibrate, diazepam, furosemide, hydrochlorothiazide, nitrazepam, oxprenolol

KEY WORDS
plasma; SPE; pharmacokinetics

REFERENCE
Riva,E.; Ferry,N.; Cometti,A.; Cuisinaud,G.; Gallo,G.G.; Sassard,J. Determination of teicoplanin in human plasma and urine by affinity and reversed-phase high-performance liquid chromatography, *J.Chromatogr.*, **1987**, *421*, 99–110.

SAMPLE
Matrix: urine
Sample preparation: Adjust pH of 1 mL urine to 7.4, add 1 mL of a homogeneous suspension of D-alanyl-D-alanine-epsilon-aminocaproyl-Sepharose resin:50 mM pH 7.4 phosphate buffer containing 200 mM NaCl 50:50, let stand at 4° overnight, place on top of a column containing 6 mL of a homogeneous suspension of D-alanyl-D-alanine-epsilon-aminocaproyl-Sepharose resin:50 mM pH 7.4 phosphate buffer containing 200 mM NaCl 50:50, wash with 6 mL 50 mM pH 7.4 phosphate buffer containing 200 mM NaCl, elute with four 3 mL portions of 1.5% aqueous ammonia, immediately neutralize the eluate with 100 mM HCl, inject an aliquot.

HPLC VARIABLES
Column: 250 × 4.6 5 μm Ultrasphere ODS
Mobile phase: Gradient. A was 25 mM pH 6.0 sodium phosphate buffer. B was MeCN:water 90:10. A:B from 84:16 to 55:45 over 2 h, wash with 49:51 for 5 min.
Column temperature: 20
Flow rate: 1.5
Detector: UV 254

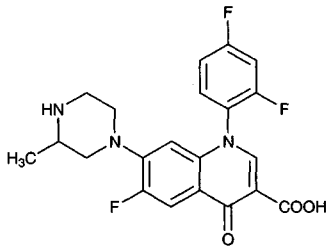
CHROMATOGRAM
Retention time: 8 (A3), 19 (A2-1), 20 (A2-2), 22 (A2-3,3a), 27 (A2-4), 28 (A2-5,5a)
Limit of detection: 0.5-5 μg/mL

OTHER SUBSTANCES
Extracted: metabolites

KEY WORDS
rat; SPE

REFERENCE
Zerilli,L.F.; Cavenaghi,L.; Bernareggi,A.; Assandri,A. Teicoplanin metabolism in rats, *Antimicrob.Agents Chemother.*, **1989**, *33*, 1791–1794.

Temafloxacin



Molecular formula: $C_{21}H_{18}F_3N_3O_3$
Molecular weight: 417.39
CAS Registry No.: 108319-06-8, 105784-61-0 (HCl)
Merck Index: 9284
Lednicer No.: 5 125

SAMPLE
Matrix: bile, blood, urine
Sample preparation: Dilute urine 1:20. Dilute bile 1:10. 500 μL Serum, diluted urine, or diluted bile + 3.2 mL dichloromethane, vortex, rotate at 20 rpm for 10 min, centrifuge at 1000 g for 10 min. Remove 3 mL of the lower organic phase and add it to 200 μL 100 mM NaOH, rotate at 20 rpm for 10 min, centrifuge at 1000 g for 10 min, inject a 20 μL aliquot of the aqueous layer.

HPLC VARIABLES
Column: 150 × 4.6 5 μm Ultrasphere C18
Mobile phase: MeCN:buffer 19:81, pH adjusted to 2 with 14.6 M phosphoric acid (Buffer was 10 mM NaH_2PO_4 containing 5 mM tetrabutylammonium bromide.)
Flow rate: 2
Injection volume: 20
Detector: F ex 275 em 450

CHROMATOGRAM
Retention time: 3
Limit of detection: 100 ng/mL (bile), 200 ng/mL (urine), 10 ng/mL (serum)

OTHER SUBSTANCES
Noninterfering: amikacin, aztreonam, carbamazepine, cephalosporins, ciprofloxacin, clavulanic acid, difloxacin, digitoxin, digoxin, fleroxacin, fosfomycin, furosemide, gentamycin, imipenem, lidocaine, netilmicin, norfloxacin, ofloxacin, pefloxacin, penicillins, phenobarbital, phenytoin, primidone, procainamide, quinidine, rifampin, salicylic acid, teicoplanin, theophylline, tobramycin, valproic acid, vancomycin

KEY WORDS
serum; human; rabbit

REFERENCE
Koechlin,C.; Jehl,F.; Linger,L.; Monteil,H. High-performance liquid chromatography for the determination of three new fluoroquinolones, fleroxacin, temafloxacin and A-64730, in biological fluids, *J.Chromatogr.*, **1989**, *491*, 379–387.

SAMPLE
Matrix: blood, tissue
Sample preparation: Homogenize (Ultra-Turrax T25) mouse lung in 1-3 mL pH 6.8 Soerensen phosphate buffer, centrifuge. Add 1 μg sparfloxacin to serum or lung homogenate supernatant, extract using a Bond Elut C2 SPE cartridge, inject a 100 μL aliquot of the extract.

HPLC VARIABLES
Column: 150 × 4.6 5 μm Ultrabase C8 (SFCC, Neuilly Plaisance, France)
Mobile phase: MeCN:MeOH:5% acetic acid 15:10:75
Flow rate: 1
Injection volume: 100
Detector: UV 280

CHROMATOGRAM
Retention time: 6
Internal standard: sparfloxacin
Limit of detection: 15 ng

KEY WORDS
serum; lung; mouse; pharmacokinetics; SPE

REFERENCE
Vallée,E.; Azoulay-Dupuis,E.; Bauchet,J.; Pocidalo,J.-J. Kinetic disposition of temafloxacin and ciprofloxacin in a murine model of pneumococcal pneumonia. Relevance for drug efficacy, *J.Pharmacol.Exp.Ther.*, **1992**, *262*, 1203–1208.

SAMPLE
Matrix: blood, urine
Sample preparation: Plasma. Mix equal volumes of plasma and 100-500 ng/mL IS in MeCN: 75 mM pH 7.4 phosphate buffer containing 0.5% sodium dodecyl sulfate 30:70, filter (Amicon Centrifree with YMT membrane) while centrifuging at less than 1000 g for 20 min, inject an aliquot of the ultrafiltrate. Alternatively, mix 400 μL plasma and 400 μL 500 mM pH 7 phosphate buffer, add 6 mL dichloromethane:EtOH 90:10, shake slowly horizontally for 10 min, centrifuge in a refrigerated centrifuge at 900 g for 10 min. Remove the organic layer and evaporate it to dryness under a stream of air at 50-60°, reconstitute the residue in mobile phase, inject an aliquot. Urine. Dilute urine 20-100 fold with mobile phase containing IS, inject an aliquot. Hydrolyze conjugates by heating urine in 1 M NaOH at 60° for 30 min, neutralize, dilute 20-100 fold with mobile phase containing IS, inject an aliquot.

HPLC VARIABLES
Column: 250 × 4.6 7 μm Adsorbosphere HS C18
Mobile phase: MeCN:water 53:47 containing 40 mM phosphoric acid, 10 mM NaH_2PO_4, 0.2% sodium dodecyl sulfate, and 5 mM N-acetylhydroxamic acid
Flow rate: 1.5
Injection volume: 50
Detector: F ex 280 em 389 (or UV 280, Antimicrob. Agents Chemother. 1991, 35, 436)

CHROMATOGRAM
Retention time: 7.5
Internal standard: 1-(4-bromophenyl)-6-fluoro-1,4-dihydro-7-(4-methyl-1-piperazinyl)-oxo-3-quinoline carboxylic acid (A57084) (8.5)
Limit of quantitation: 1 ng/mL (extraction), 10 ng/mL (ultrafiltrate)

OTHER SUBSTANCES
Extracted: metabolites

KEY WORDS
plasma; ultrafiltrate; pharmacokinetics

REFERENCE
Granneman,G.R.; Varga,L.L. High-performance liquid chromatographic procedures for the determination of temafloxacin in biological matrices, *J.Chromatogr.*, **1991**, *568*, 197–206.

SAMPLE
Matrix: blood, urine
Sample preparation: 1 mL Serum or 500 μL urine + 500 μL 1 M ammonium acetate + 5 mL dichloromethane, shake vigorously for 10 min, centrifuge at 1500 g for 5 min. Remove the organic layer and evaporate it to dryness under a stream of nitrogen at 40°, reconstitute the residue in 500 μL MeOH:thionyl chloride 90:10, heat at 60° for 30 min, evaporate to dryness under a stream of nitrogen at 40°, add 500 μL 500 mM sulfuric acid, add 1 mL hexane, shake vigorously for 10 min, centrifuge for 5 min. Remove the aqueous layer and add it to 1 mL 2 M sodium carbonate and 3 mL ether, shake vigorously for 10 min, centrifuge for 5 min. Remove the organic layer and evaporate it to dryness under a stream of nitrogen at 40°, reconstitute the residue in 20 μL 2 mg/mL (S)-(-)-N-1-(2-naphthylsulfonyl)-2- pyrrolidinecarbonyl chloride in dichloromethane, add 5 μL triethylamine, let stand at room temperature for 10 min. Evaporate to dryness under a stream of nitrogen, reconstitute the residue in 500 μL dichloromethane, inject a 50 μL aliquot. (No details of synthesis of chiral reagent given in paper.)

HPLC VARIABLES
Column: 150 × 4.6 Zorbax SIL
Mobile phase: Hexane:methyl acetate:MeOH:aqueous ammonia 150:100:10:1
Flow rate: 0.8
Injection volume: 50
Detector: UV 280

CHROMATOGRAM
Retention time: 18.5 (S), 19.5 (R)
Limit of detection: 5 ng/mL

KEY WORDS
serum; chiral; pharmacokinetics; derivatization; normal phase

REFERENCE
Matsuoka,M.; Banno,K.; Sato,T. Analytical chiral separation of a new quinolone compound in biological fluids by high-performance liquid chromatography, *J.Chromatogr.B*, **1996**, *676*, 117–124.

SAMPLE
Matrix: bulk
Sample preparation: Dissolve 100 mg bulk drug in 25 mL MeCN:water 50:50 containing 1 mL 1 M NaOH, add 15 mL 20 mM KH_2PO_4 adjusted to pH 2.4 with orthophosphoric acid, make up to 50 mL with MeCN:water 50:50, inject a 50 μL aliquot.

HPLC VARIABLES
Column: 150 × 4.6 5 μm Nucleosil C18
Mobile phase: Gradient. MeCN:THF:buffer from 5:5:90 to 5:60:35 over 50 min, maintain at 5:60:35 for 10 min, return to initial conditions over 5 min, re-equilibrate for 15 min. (Buffer was 20 mM KH_2PO_4 adjusted to pH 2.4 with orthophosphoric acid.)
Flow rate: 1
Injection volume: 50
Detector: UV 325

CHROMATOGRAM
Retention time: 11
Limit of detection: 0.05% (of temafloxacin)

OTHER SUBSTANCES
Simultaneous: degradation products, impurities

REFERENCE
Elrod,L.,Jr.; Linton,C.L.; Shelat,B.P.; Wong,C.F. Determination of minor impurities in temafloxacin hydrochloride by high-performance liquid chromatography, *J.Chromatogr.*, **1990**, *519*, 125–136.

SAMPLE
Matrix: cells
Sample preparation: Incubate cells in 2 mL 100 mM pH 3.0 glycine-HCl buffer for 2 h at room temperature, centrifuge at 5600 g for 5 min, inject an aliquot.

HPLC VARIABLES
Column: Bondapak C18
Mobile phase: MeCN:25 mM phosphoric acid adjusted to pH 3.0 with tetrabutylammonium hydroxide 25:75
Flow rate: 1.5
Detector: F ex 340 em 425

OTHER SUBSTANCES
Also analyzed: ciprofloxacin, fleroxacin, lomefloxacin, norfloxacin, ofloxacin

REFERENCE
Pascual,A.; Garcia,I.; Conejo,M.C.; Perea,E.J. Fluorometric and high-performance liquid chromatographic measurement of quinolone uptake by human neutrophils, *Eur.J.Clin.Microbiol.Infect.Dis.*, **1991**, *10*, 969–971.

SAMPLE
Matrix: hair
Sample preparation: Hair. Cut 10 hairs into 2 mm sections, weigh, add 1 mL 1 M NaOH, heat at 60° for 1 h, cool, add 500 μL 2 M HCl, add 1 mL 200 mM pH 7.0 phosphate buffer, add 500 μL 4-20 ng/mL IS in MeOH, add 5 mL chloroform, shake for 30 min, centrifuge at 2000 rpm for 10 min. Remove 4 mL of the organic layer and evaporate it to dryness by aspiration at 40° for 30 min, reconstitute the residue in 200-500 μL mobile phase, inject a 100 μL aliquot.

HPLC VARIABLES
Column: 150 × 4.6 TSKgel ODS-80TM (Tosoh)
Mobile phase: MeCN:50 mM citric acid:1 M ammonium acetate 22:78:1
Flow rate: 1
Injection volume: 100
Detector: F ex 280 em 406

CHROMATOGRAM
Retention time: 12
Internal standard: 1-(4-bromophenyl)-6-fluoro-1,4-dihydro-7-(4-methyl-1-piperazinyl)-oxo-3-quinoline carboxylic acid (A57084) (14)
Limit of detection: 0.5 ng/mL

REFERENCE
Uematsu,T.; Kondo,K.; Yano,S.; Yamaguchi,T.; Umemura,K.; Nakashima,M. Measurement of temafloxacin in human scalp hair as an index of drug exposure, *J.Pharm.Sci.*, **1994**, *83*, 42–45.

SAMPLE
Matrix: solutions
Sample preparation: Prepare a 450 μg/mL solution in MeCN:water 50:50. 5 mL Solution + 5 mL THF + 200 molar excess of acetic anhydride + 3 molar excess of 1 M NaOH, sonicate for 15 min, add 15 mL mobile phase, sonicate for 15 min, cool to room temperature, make up to 50 mL with mobile phase, inject a 50 μL aliquot.

HPLC VARIABLES
Column: 150 × 4.6 5 μm Nucleosil C18

Mobile phase: MeCN:buffer 35:65 (Buffer was prepared by mixing equal volumes of 20 mM citric acid and 20 mM sodium citrate, pH adjusted to 2.4 with perchloric acid.)
Flow rate: 1
Injection volume: 50
Detector: UV 280

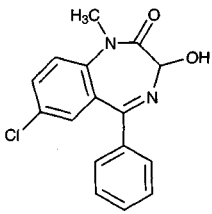
CHROMATOGRAM
Retention time: 21.4

OTHER SUBSTANCES
Simultaneous: ciprofloxacin, norfloxacin, sarafloxacin

KEY WORDS
derivatization

REFERENCE
Morley,J.A.; Elrod,L.,Jr. Determination of fluoroquinolone antibacterials as N-Acyl derivatives, *Chromatographia*, **1993**, *37*, 295–299.

Temazepam



Molecular formula: $C_{16}H_{13}ClN_2O_2$
Molecular weight: 300.74
CAS Registry No.: 846-50-4
Merck Index: 9285
Lednicer No.: 2 402

SAMPLE
Matrix: bile, blood, gastric contents, tissue, urine
Sample preparation: Chop 5-g tissue and homogenize (Ultra Turrax T25) at 8500, 9500, 13500, 20500, and 24000 rpm for 1 min each. Add homogenate to 20 mL water. Dilute blood, urine, gastric contents, and bile four times with water. Mix 4 mL sample with 10 μL 1 mg/mL prazepam and 1 mL pH 7.4 phosphate buffer, vortex briefly, add 4 mL diethyl ether and mix for 15 min (Spiramix 10, Denley, UK). Separate the organic layer, add 4 mL diethyl ether to extraction sample, mix. Evaporate combined organic layers to dryness under a stream of dry air at 50°. Purify extracts by partition between 1 mL MeCN and 2 mL heptane, separate MeCN layer, evaporate it to dryness, reconstitute the residue in 100 μL MeOH and inject a 20 μL aliquot of the solution.

HPLC VARIABLES
Guard column: 20 × 4.6 5 μm Apex II ODS
Column: 150 × 4.6 5 μm Apex II ODS
Mobile phase: MeCN:MeOH:10 mM phosphoric acid:10 mM Na_2HPO_4 40:20:36:4
Flow rate: 1
Injection volume: 20
Detector: UV 240

CHROMATOGRAM
Retention time: 4.8
Internal standard: prazepam (14.5)
Limit of quantitation: 100 ng/mL

OTHER SUBSTANCES
Extracted: diazepam, nitrazepam, oxazepam

KEY WORDS
liver; lung; muscle; urine; pericardial fluid

REFERENCE
Pounder,D.J.; Adams,E.; Fuke,C.; Langford,A.M. Site to site variability of postmortem drug concentrations in liver and lung, *J.Forensic Sci.*, **1996**, *41*, 927–932.

SAMPLE
Matrix: blood
Sample preparation: 200 mg Extrelut + 400 µL blood + 100 µL 50 µg/mL IS in MeOH, mix, let dry at room temperature for 1-2 h. Add to a 30 × 4.6 stainless steel extraction column, extract with carbon dioxide:ethyl acetate 95:5 at 2 mL/min, 65°, and 300 psi. for 10 min, collect by expansion into MeOH. Dry the collected extract at 65° under nitrogen. Reconstitute the residue in 50 µL mobile phase. Inject a 20 µL aliquot. Condition ca. 10 g Extrelut in a 10 mL plastic syringe with dichloromethane. Add 250 µL 5% ammonia to the top. Mix 900 µL blood with 100 µL 50 µg/mL IS in MeOH. Add 1 mL pH 4 phosphate buffer and 250 µL 5% ammonia solution, mix thoroughly, add to the extraction column. After 5 min elute with diethyl ether under the influence of gravity. Collect 8 mL eluate, evaporate to dryness at 65° under nitrogen. Reconstitute the residue in 180 µL mobile phase, inject a 20 µL aliquot.

HPLC VARIABLES
Guard column: 20 × 4.6 5 µm Hypersil ODS
Column: 250 × 4.6 5 µm Hypersil ODS
Mobile phase: MeOH:Na_2HPO_4 70:30
Flow rate: 1
Injection volume: 20
Detector: UV 254

CHROMATOGRAM
Retention time: 3
Internal standard: prazepam (11.5)

OTHER SUBSTANCES
Also analyzed: diazepam, chlordiazepoxide, nordiazepam, oxazepam

KEY WORDS
SFE; SPE; whole blood

REFERENCE
Scott,K.S.; Oliver,J.S. Development of a supercritical fluid extraction method for the determination of temazepam in whole blood, *J.Anal.Toxicol.*, **1997**, *21*, 297–300.

SAMPLE
Matrix: blood, urine
Sample preparation: Add 1 mL whole blood or urine to Toxi-Tube A (Toxi-Lab, Irvine CA), add 3 mL water, mix by gentle inversion for 5 min, centrifuge at 1500 g for 5 min. Remove the organic layer and evaporate it to dryness under a stream of nitrogen at 40°, reconstitute the residue with 50 µL MeCN:water 50:50, vortex for 10 s, centrifuge at 7500 g for 2 min, inject a 10 (urine) or 30 (blood) µL aliquot. (The detector wavelength shown is the wavelength of maximum absorbance. This will not necessarily be the optimal wavelength for the separation. Multiple wavelengths from 200-350 nm can be scanned using a diode-array detector. Otherwise, 220 nm may be a reasonable choice for initial work. Matrix may interfere.)

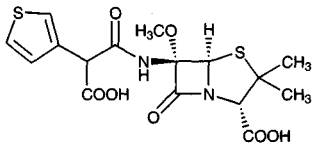
HPLC VARIABLES
Guard column: 20 mm long Symmetry C18
Column: 250 × 4.6 5 µm Symmetry C8 (Waters)
Mobile phase: Gradient. A was 50 mM pH 3.8 sodium phosphate buffer. B was MeCN. A:B 85:15 for 6.5 min, 65:35 for 18.5 min, 20:80 for 3 min (step gradient), re-equilibrate at initial conditions for 7 min.
Column temperature: 30
Flow rate: 1 for 6.5 min, to 1.5 over 18.5 min, maintain at 1.5 for 3 min (re-equilibrate at 1.5 mL/min)
Injection volume: 10-30
Detector: UV 200.5

CHROMATOGRAM
Retention time: 18.562

KEY WORDS
whole blood

REFERENCE
Gaillard,Y.; Pépin,G. Use of high-performance liquid chromatography with photodiode-array UV detection for the creation of a 600-compound library. Application to forensic toxicology, *J.Chromatogr.A*, **1997**, *763*, 149–163.

Temocillin



Molecular formula: $C_{16}H_{18}N_2O_7S_2$
Molecular weight: 414.46
CAS Registry No.: 66148-78-5
Merck Index: 9288
Lednicer No.: 4 178

SAMPLE
Matrix: blood
Sample preparation: 350 μL Serum + 150 μL water + 250 μL 400 mM HCl + 3.5 mL chloroform: n-amyl alcohol (3:1), mix for 5 min, centrifuge for 5 min. Remove the organic layer and add it to 350 μL 10 mM pH 7.0 phosphate buffer, mix for 5 min, centrifuge for 5 min, inject a 20 μL aliquot of the upper aqueous layer.

HPLC VARIABLES
Column: 300 × 3.9 10 μm μBondapak C18
Mobile phase: MeOH:buffer 15:85 (Buffer was 100 mM ammonium acetate adjusted to pH 4.0 with glacial acetic acid.)
Flow rate: 1.8
Injection volume: 20
Detector: UV 242

CHROMATOGRAM
Retention time: 5.4
Internal standard: temocillin

OTHER SUBSTANCES
Extracted: cefoxitin, cefuroxime, cephalothin, ticarcillin
Noninterfering: acetaminophen, acetazolamide, allopurinol, amikacin, ampicillin, azlocillin, caffeine, cefamandole, cefoperazone, cefotaxime, cefsulodin, ceftazidime, ceftizoxime, chloramphenicol, chlorpromazine, clindamycin, dicloxacillin, 5-fluorocytosine, flurazepam, gentamicin, methicillin, metronidazole, mezlocillin, moxalactam, nafcillin, penicillin, phenobarbital, piperacillin, procainamide, rifampin, sulfamethoxazole, theophylline, thienamycin, tobramycin, trimethoprim, vancomycin

KEY WORDS
serum; temocillin is IS

REFERENCE
Shull,V.H.; Dick,J.D. Determination of ticarcillin levels in serum by high-pressure liquid chromatography, *Antimicrob.Agents Chemother.*, **1985**, *28*, 597–600.

SAMPLE
Matrix: bulk

Sample preparation: Prepare a 300 μg/mL solution in 100 mM pH 7.0 phosphate buffer, inject a 20 μL aliquot.

HPLC VARIABLES
Column: 300 × 3.9 μBondapak C18
Mobile phase: MeOH:buffer 10:90 (Buffer was 15.6 g $NaH_2PO_4.2H_2O$ in 900 mL water, adjust pH to 7.0 with NaOH, make up to 1 L with water.)
Flow rate: 2
Injection volume: 20
Detector: UV 230

CHROMATOGRAM
Retention time: 6.0 (R), 7.5 (S)

OTHER SUBSTANCES
Simultaneous: isomers, diastereomers, impurities, degradation products, ticarcillin

REFERENCE
Bird,A.E.; Charsley,C.H.; Jennings,K.R.; Marshall,A.C. High-performance liquid chromatographic assay of temocillin and epimerisation of its diastereoisomers, *Analyst*, **1984**, *109*, 1209–1212.

SAMPLE
Matrix: solutions

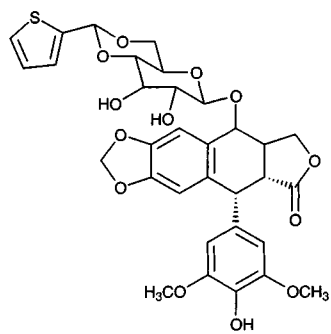
HPLC VARIABLES
Column: 300 × 3.9 μBondapak C18
Mobile phase: MeOH:100 mM pH 6.5 potassium phosphate 10:90
Flow rate: 2
Injection volume: 50
Detector: UV 254

KEY WORDS
radiolabelled compounds

REFERENCE
Morecombe,D.J. High-efficiency preparative-scale reversed-phase high-performance liquid chromatographic purification of 14C-labelled antibiotics, *J.Chromatogr.*, **1987**, *389*, 389–395.

Teniposide

Molecular formula: $C_{32}H_{32}O_{13}S$
Molecular weight: 656.66
CAS Registry No.: 29767-20-2
Merck Index: 9291



SAMPLE
Matrix: blood
Sample preparation: Filter 1 mL plasma (Centrifree micropartition device, molecular mass cut-off 30000, Amicon, USA) using a 33° fixed angle centrifuge (Beckman ModelG56R) at 2000 g and 25° for 30 min. Add 1 mL chloroform to the ultrafiltrate (Caution! Chloroform is a carcinogen!), agitate slowly for 20 min, centrifuge at 1000 g for 5 min. Evaporate the organic phase to dryness under vacuum at 40°, dissolve the dry extract in 50 μL MeOH, inject a 25 μL aliquot.

HPLC VARIABLES
Column: 300 × 3.9 10 μm 125 Å μBondapak Phenyl (Waters)
Mobile phase: MeCN:water:glacial acid 35:64:1
Flow rate: 1
Injection volume: 25
Detector: F ex 288 em 328

CHROMATOGRAM
Retention time: 18
Internal standard: teniposide

OTHER SUBSTANCES
Extracted: etoposide
Noninterfering: alizapride, doxorubicin, furosemide, idarubicin, ranitidine, vinblastine, vinorelbine

KEY WORDS
plasma; ultrafiltrate; teniposide is IS

REFERENCE
Robieux,I.; Aita,P.; Sorio,R.; Toffoli,G.; Boiocchi,M. Determination of unbound etoposide concentration in ultrafiltered plasma by high-performance liquid chromatography with fluorimetric detection, *J.Chromatogr.B*, **1996**, *686*, 35–41.

SAMPLE
Matrix: blood
Sample preparation: 1 mL Plasma + 10 μL 1 mg/mL etoposide in MeOH + 5 mL chloroform, rock gently for 15 min, centrifuge. Remove 4.5 mL of the organic layer and evaporate it to dryness under a stream of nitrogen at 40°, dissolve residue in 50 μL MeOH, vortex, centrifuge for 5-10 min, inject a 20 μL aliquot.

HPLC VARIABLES
Column: 300 × 3.9 10 μm μBondapak C18
Mobile phase: MeOH:water 60:40
Flow rate: 1-1.2
Injection volume: 20
Detector: UV 254

CHROMATOGRAM
Retention time: 7.5
Internal standard: etoposide (5)
Limit of quantitation: 500 ng/mL

KEY WORDS
plasma

REFERENCE
Strife,R.J.; Jardine,I.; Colvin,M. Analysis of the anticancer drugs VP 16-213 and VM 26 and their metabolites by high-performance liquid chromatography, *J.Chromatogr.*, **1980**, *182*, 211–220.

SAMPLE
Matrix: blood
Sample preparation: 1 mL Plasma + 10 μL 100 μg/mL etoposide in MeOH + 5 mL chloroform, rock gently for 15 min, centrifuge. Remove 4.5 mL of the organic layer and evaporate it to dryness under a stream of nitrogen at 40°, dissolve residue in 50 μL MeOH, vortex, centrifuge for 5-10 min, inject a 25 μL aliquot.

HPLC VARIABLES
Guard column: 70 × 2.1 30 μm Co:Pell (Whatman)
Column: 300 × 3.9 10 μm μBondapak C18
Mobile phase: MeOH:water 60:40

Flow rate: 1
Injection volume: 25
Detector: F ex 215 em 328

CHROMATOGRAM
Retention time: 8
Internal standard: etoposide (5.5)
Limit of detection: 25 ng/mL
Limit of quantitation: 50 ng/mL

KEY WORDS
plasma

REFERENCE
Strife,R.J.; Jardine,I.; Colvin,M. Analysis of the anticancer drugs etoposide (VP 16-213) and teniposide (VM 26) by high-performance liquid chromatography with fluorescence detection, *J.Chromatogr.*, **1981**, *224*, 168–174.

SAMPLE
Matrix: blood
Sample preparation: Mix plasma or serum with an equal volume of proteinase K, let stand for 10 min. (Alternatively, heat serum or plasma with an equal volume of 1 mg/mL subtilisin A at 50° for 15 min.) Inject 1.6 mL hydrolyzed blood or filtered serum on to column A with mobile phase A at 1 mL/min, backflush column A with mobile phase A to waste for 2 min at 2 mL/min, backflush the contents of column A on to column B with mobile phase B, after 30 s remove column A from the circuit, elute column B with mobile phase B, monitor the effluent from column B. (Clean column A by backflushing with MeOH at 2 mL/min for 3 min then forward flush with water at 1 mL/min for 2 min.)

HPLC VARIABLES
Column: A 2 × 4.6 10 μm PRP-1 divinylbenzene-styrene copolymer (Hamilton); B 125 × 4 10 μm LiChrosorb RP-18
Mobile phase: A water; B MeOH:water 55:45
Flow rate: A 1-2; B 1
Injection volume: 1600
Detector: F ex 230 em 328 following post-column extraction. The column effluent mixed with dichloroethane pumped at 0.6 mL/min, the mixture flowed through a 2 mm i.d. glass reactor (Technicon) to a phase separator (Technicon (J.Chromatogr. 1979, 185, 473)) with a PTFE insert and 0.3 mL/min of the organic phase flowed through the detector.

CHROMATOGRAM
Retention time: 8

OTHER SUBSTANCES
Extracted: etoposide

KEY WORDS
column-switching; post-column extraction; plasma; serum

REFERENCE
Werkhoven-Goewie,C.E.; Brinkman,U.A.T.; Frei,R.W.; de Ruiter,C.; de Vries,J. Automated liquid chromatographic analysis of the anti-tumorigenic drugs etoposide (VP 16-213) and teniposide (VM 26), *J.Chromatogr.*, **1983**, *276*, 349–357.

SAMPLE
Matrix: blood
Sample preparation: 1 mL Plasma + 5 μg etoposide + 3 mL chloroform, shake for 10 min, centrifuge at 1500 g for 10 min, repeat extraction. Combine the organic layers and evaporate a 5 mL aliquot to dryness under a stream of nitrogen at 50°, reconstitute the residue in 200 μL mobile phase, inject a 20 μL aliquot.

HPLC VARIABLES
Column: 300 × 3.9 10 μm μBondapak C18
Mobile phase: MeOH:250 mM ammonium acetate:acetic acid 54:45:1
Flow rate: 1.5
Injection volume: 20
Detector: E, Bioanalytical Systems LC4, TL5 glassy carbon electrode, + 900 mV, Ag/AgCl reference electrode

CHROMATOGRAM
Retention time: 8.3
Internal standard: etoposide (4.1)
Limit of detection: 10 ng/mL
Limit of quantitation: 20 ng/mL

KEY WORDS
plasma; pharmacokinetics

REFERENCE
Canal,P.; Michel,C.; Bugat,R.; Soula,G.; Carton,M. Quantification of teniposide in human serum by high-performance liquid chromatography with electrochemical detection, *J.Chromatogr.*, **1986**, *375*, 451–456.

SAMPLE
Matrix: blood
Sample preparation: 450 μL Plasma + 50 μL 380 mM sodium dodecyl sulfate in 59 mM pH 7 sodium phosphate buffer + 5 μL 500 ng/mL etoposide in MeOH, sonicate for 5 min. Inject a 100 μL aliquot onto column A with mobile phase A, elute with mobile phase A for 7.5 min, elute the contents of column A onto column B with mobile phase B for 1 min. After 1 min remove column A from the circuit and re-equilibrate it with mobile phase A for 1.5 min, monitor the effluent from column B.

HPLC VARIABLES
Column: A 10 × 2.1 40 μm Chromsep C18 (Chrompack); B 300 × 4.6 10 μm μBondapak phenyl
Mobile phase: A 10 mM pH 7.0 sodium phosphate; B MeOH:10 mM pH 7.0 sodium phosphate buffer 55:45
Flow rate: A 0.4; B 1
Injection volume: 100
Detector: E, +500 mV vs Ag/AgCl or UV 254

CHROMATOGRAM
Retention time: 6.4
Internal standard: etoposide (4.2)
Limit of detection: 20 ng/mL (electrochemical), 150 ng/mL (UV)

KEY WORDS
plasma; column-switching

REFERENCE
van Opstal,M.A.J.; van der Horst,F.A.L.; Holthuis,J.J.M.; Van Bennekom,W.P.; Bult,A. Automated reversed-phase chromatographic analysis of etoposide and teniposide in plasma by using on-line surfactant-mediated sample clean-up and column-switching, *J.Chromatogr.*, **1989**, *495*, 139–151.

SAMPLE
Matrix: blood
Sample preparation: 1 mL Plasma + 50 μL 100 μg/mL etoposide in MeOH, vortex, add 2 mL dichloroethane, shake thoroughly for 1 min, centrifuge at 3000 g for 5 min. Remove 1. 5 mL of the organic layer and evaporate it to dryness under a stream of nitrogen, reconstitute the residue in 150 μL MeOH:water 70:30, sonicate for 6 min, inject a 10 μL aliquot.

HPLC VARIABLES
Guard column: 10 × 4.6 10 μm LiChrosorb C18
Column: 100 × 4.6 10 μm Novapak phenyl

Mobile phase: MeOH:10 mM pH 7.0 phosphate buffer 55:45
Flow rate: 0.7
Injection volume: 10
Detector: E, Metrohm Model 641 VA, EA 286/1 glassy carbon electrode + 500 mV, stainless-steel auxiliary electrode, Ag/AgCl reference electrode

CHROMATOGRAM
Retention time: 7
Internal standard: etoposide (4.5)
Limit of detection: 10 ng/mL

KEY WORDS
plasma

REFERENCE
van der Horst,F.A.L.; van Opstal,M.A.J.; Teeuwsen,J.; Post,M.H.; Holthuis,J.J.M.; Brinkman,U.A.T. Comparative study on the determination of the anti-neoplastic drug teniposide in plasma using micellar liquid chromatography and surfactant-mediated plasma clean-up, *J.Chromatogr.*, **1991**, *567*, 161–174.

SAMPLE
Matrix: blood
Sample preparation: Sonicate 50 million leukemic cells in 1 mL phosphate buffered saline. 500 μL Plasma or 1 mL sonicated cells + 2 mL chloroform, mix. Remove the organic layer and evaporate it to dryness under a stream of nitrogen, reconstitute the residue in 200 μL MeOH: water 50:50, sonicate for 5 min, inject a 100 μL aliquot. To measure non-protein-bound etoposide filter (Amicon Centrifree) while centrifuging at 20°, inject a 100-200 μL aliquot of the ultrafiltrate.

HPLC VARIABLES
Column: 250 × 4.6 5 μm Spherisorb Phenyl
Mobile phase: MeOH:water:acetic acid 45:54:1
Flow rate: 1
Injection volume: 100-200
Detector: F ex 220 em 330

CHROMATOGRAM
Retention time: 9.3
Internal standard: teniposide

OTHER SUBSTANCES
Extracted: etoposide, cis-etoposide

KEY WORDS
plasma; cells; ultrafiltrate; teniposide is IS

REFERENCE
Liliemark,E.; Petterson,B.; Peterson,C.; Liliemark,J. High-performance liquid chromatography with fluorometric detection for monitoring of etoposide and its *cis*-isomer in plasma and leukaemic cells, *J.Chromatogr.B*, **1995**, *669*, 311–317.

SAMPLE
Matrix: blood
Sample preparation: 400 μL Plasma + 1.6 mL diethyl ether, vortex, centrifuge at 1000 g for 2 min. Remove the organic layer and evaporate it to dryness under a stream of air at 30°, reconstitute the residue in 200 μL mobile phase, inject a 100 μL aliquot.

HPLC VARIABLES
Column: 250 × 4 5 μm LiChrospher 100 RP-18
Mobile phase: MeCN:MeOH:buffer 30:20:50 (Buffer was 20 g/L NaH_2PO_4 containing 0.8 g/L heptanesulfonic acid, pH adjusted to 3.0 with orthophosphoric acid.)
Flow rate: 1

Injection volume: 100
Detector: E, Environmental Sciences Coulochem 5100 A, guard cell +0.90 V (before injector), clean-up cell +0.40 V, detection cell +0.90 V

CHROMATOGRAM
Retention time: 10.6
Internal standard: teniposide

OTHER SUBSTANCES
Extracted: vinorelbine

KEY WORDS
plasma; rabbit; teniposide is IS

REFERENCE
Mouchard-Delmas,C.; Gourdier,B.; Vistelle,R. Determination of vinorelbine in rabbit plasma by high-performance liquid chromatography with coulometric detection, *J.Chromatogr.B*, **1995**, *663*, 390–394.

SAMPLE
Matrix: blood, CSF, urine
Sample preparation: 500 μL Plasma, urine, or CSF + 500 μL saturated ammonium sulfate + 4 mL ethyl acetate + 10 μL 100 μg/mL etoposide, vortex for 5 min, centrifuge at 3000 rpm for 15 min, repeat the extraction. Combine the organic layers and evaporate them to dryness under a stream of nitrogen, reconstitute the residue in 200 μL MeOH, inject an aliquot.

HPLC VARIABLES
Guard column: 10 μm μBondapak phenyl
Column: 250 × 4.6 10 μm μBondapak phenyl
Mobile phase: MeCN:water:acetic acid 30:68:2
Flow rate: 1
Injection volume: 50
Detector: UV 284 or E, Bioanalytical Systems LC-4A, 0.75 V

CHROMATOGRAM
Retention time: 18
Internal standard: etoposide (7)
Limit of detection: 20 ng/mL
Limit of quantitation: 50 ng/mL

OTHER SUBSTANCES
Extracted: metabolites

KEY WORDS
plasma; pharmacokinetics

REFERENCE
Sinkule,J.A.; Evans,W.E. High-performance liquid chromatographic analysis of the semisynthetic epipodophyllotoxins teniposide and etoposide using electrochemical detection, *J.Pharm.Sci.*, **1984**, *73*, 164–168.

SAMPLE
Matrix: solutions
Sample preparation: Prepare a solution in MeCN:water:650 mM pH 4.0 sodium citrate 40:60:0.4, inject a 30 μL aliquot.

HPLC VARIABLES
Guard column: 10 × 3.9 10 μm μBondapak phenyl
Column: 150 × 3.9 10 μm μBondapak phenyl
Mobile phase: MeCN:water:650 mM pH 4.0 sodium citrate 35:57.3:7.7. When run is over wash with MeCN:water:650 mM pH 4.0 sodium citrate 70:27.3:7.7 for 4 min, re-equilibrate with initial mobile phase for 9 min.

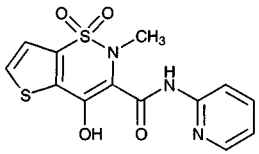
Flow rate: 2
Injection volume: 30
Detector: E, ESA 5100A detector, Model 5020 guard cell between pump and autosampler +0.7 V, Model 5011 dual electrode analytical cell, upstream (screening) electrode +0.2 V, downstream electrode +0.45 V against Ag/AgCl

CHROMATOGRAM
Retention time: 5.8

OTHER SUBSTANCES
Simultaneous: etoposide

REFERENCE
Eisenberg,E.J.; Eickhoff,W.M. Determination of etoposide in blood by liquid chromatography with electrochemical detection, *J.Chromatogr.*, **1993**, *621*, 110–114.

Tenoxicam



Molecular formula: $C_{13}H_{11}N_3O_4S_2$
Molecular weight: 337.38
CAS Registry No.: 59804-37-4
Merck Index: 9293
Lednicer No.: 4 173

SAMPLE
Matrix: blood
Sample preparation: 1 mL Plasma + 1 mL pH 2 phosphate buffer + 10 mL diethyl ether, vortex for 1 min, centrifuge at 1300 g for 5 min. Evaporate the organic layer to dryness under a stream of nitrogen. Reconstitute the residue in 100 μL 10 mM HCl in MeOH, inject a 40 μL aliquot.

HPLC VARIABLES
Column: 300 × 3.9 10 μm μBondapak ODS
Mobile phase: MeOH:10 mM pH 2 phosphate buffer 45:55
Flow rate: 1.5
Injection volume: 40
Detector: UV 361

CHROMATOGRAM
Retention time: 5.81
Internal standard: tenoxicam

KEY WORDS
plasma; rat; tenoxicam is IS

REFERENCE
Amanlou,M.; Dehpour,A.R. Rapid method for the determination of piroxicam in rat plasma using high-performance liquid chromatography, *J.Chromatogr.B*, **1997**, *696*, 317–319.

SAMPLE
Matrix: blood
Sample preparation: Mix plasma with 800 μL 5 mM pH 4 phosphate buffer. Centrifuge at 2500 rpm for 5 min. Add the supernatant to Extrelut-1 cartridge (Merck, Darmstadt, Germany). Elute with 10 mL dichloromethane, dry eluate under a stream of nitrogen at 35°, dissolve the residue in 300 μL mobile phase, centrifuge and inject 100 μL aliquot of the supernatant.

HPLC VARIABLES
Column: 250 × 4.6 5 μm Hypersil ODS
Mobile phase: MeOH:50 mM pH 6 phosphate buffer 50:50
Column temperature: 35
Flow rate: 1.3
Injection volume: 100
Detector: UV 371

CHROMATOGRAM
Retention time: 2.2
Internal standard: tenoxicam

KEY WORDS
plasma; tenoxicam is IS

REFERENCE
Bareggi,S.R.; Gambaro,V.; Valenti,M.; Benvenuti,C. Absorption of oral lornoxicam in healthy volunteers using a granular formulation in comparison with standard tablets, *Arzneimittelforschung*, **1997**, *47*, 755–757.

SAMPLE
Matrix: blood
Sample preparation: 1 mL Plasma + 1 mL 1 M HCl + 1 mL water + 100 μL 2 μg/mL piroxicam + 100 μL MeOH + 5 mL dichloromethane, mix for 5 min, centrifuge at 1800 g for 10 min. Remove the organic layer and evaporate it to dryness under a stream of nitrogen at 30°, reconstitute the residue in 100 μL mobile phase, inject a 40 μL aliquot.

HPLC VARIABLES
Column: 125 × 4 5 μm LiChrospher 100 RP-18
Mobile phase: MeOH:100 mM pH 7.4 phosphate buffer 40:60
Flow rate: 1.1
Injection volume: 40
Detector: UV 355

CHROMATOGRAM
Retention time: 3.4
Internal standard: piroxicam (4.5)
Limit of detection: 5 ng/mL
Limit of quantitation: 10 ng/mL

KEY WORDS
plasma

REFERENCE
Múnera-Jaramillo,M.I.; Botero-Garcés,S. Determination of tenoxicam in plasma by high-performance liquid chromatography, *J.Chromatogr.*, **1993**, *616*, 349–352.

SAMPLE
Matrix: blood
Sample preparation: 1 mL Plasma + 10 μg ketorolac in water + 100 μL 5% zinc sulfate in water, vortex for 2 min, add 440 μL buffer, vortex for 1 min, centrifuge at 2000 g for 10 min, inject a 100 μL aliquot of the supernatant. (Buffer was 100 mM NaH_2PO_4 and 10 mM sodium lauryl sulfate, pH adjusted to 2.8 with phosphoric acid.)

HPLC VARIABLES
Column: 250 × 4.6 5 μm Nucleosil C18
Mobile phase: MeCN:water 35:65 containing 10 mM NaH_2PO_4 and 1 mM sodium lauryl sulfate, pH adjusted to 2.8 with phosphoric acid
Flow rate: 1.5
Injection volume: 100
Detector: UV 355

CHROMATOGRAM
Retention time: 4.6
Internal standard: ketorolac (10.3)
Limit of detection: 40 ng/mL

KEY WORDS
plasma; protect from light; pharmacokinetics

REFERENCE
Mason,J.L.; Hobbs,G.J. Simple method for the analysis of tenoxicam in human plasma using high-performance liquid chromatography, *J.Chromatogr.B*, **1995**, *665*, 410–415.

SAMPLE
Matrix: blood
Sample preparation: 2 mL Whole blood or plasma + 2 mL buffer + 5 mL chloroform:isopropanol: n-heptane 60:14:26, shake gently horizontally for 10 min, centrifuge at 2800 g for 10 min. Remove the lower organic layer and evaporate it to dryness under vacuum at 45°, reconstitute the residue in 100 μL mobile phase, centrifuge at 2800 g for 5 min, inject a 50 μL aliquot of the supernatant. (Buffer was saturated ammonium chloride solution 25% diluted with water, adjusted to pH 9.5 with 25% ammonia solution.)

HPLC VARIABLES
Column: 300 × 3.9 4 μm NovaPack C18
Mobile phase: MeOH:THF:buffer 65:5:30 (Buffer was 0.68 g/L (10 mM (sic)) KH_2PO_4 adjusted to pH 2.6 with concentrated orthophosphoric acid.) (At the end of each session wash the column with water for 1 h and MeOH for 1 h, re-equilibrate for 30 min.)
Column temperature: 30
Flow rate: 0.8
Injection volume: 50
Detector: UV 376

CHROMATOGRAM
Retention time: 2.99
Limit of detection: <120 ng/mL

KEY WORDS
whole blood; plasma; interferences may occur–compounds (all of which are extracted) elute in this order tenoxicam; iproniazid; methocarbamol; methotrexate; caffeine; nialamide; colchicine; cytarabine; benzoylecgonine; acetaminophen; diazoxide; dacarbazine; sulfinpyrazole; flumazenil; sulpride; morphine; atenolol; toloxatone; terbutaline; albuterol; phenobarbital; ranitidine; tiapride; phenol; chlormezanone; aspirin; metformin; ritodrine; codeine; sultopride; amisulpride; naltrexone; lisinopril; benzocaine; nizatidine; nalorphine; mephenesin; naloxone; sotalol; carteolol; procainamide; carbamazepine; bromazepam; nalbuphine; nadolol; procarbazine; dihydralazine; omeprazole; strychnine; acebutolol; glutethimide; chlorpropamide; glipizide; triazolam; prazosin; flunitrazepam; clonazepam; metoclopramide; melphalan; estazolam; tolbutamide; ephedrine; clonidine; pindolol; clobazam; minoxidil; disopyramide; nitrazepam; dextromethorphan; tofisopam; zopiclone; debrisoquine; sulindac; alprazolam; cycloguanil; lorazepam; methaqualone; ketamine; piroxicam; metoprolol; nifedipine; quinine; mephentermine; prilocaine; pentazocine; oxazepam; tiaprofenic acid; quinidine; celiprolol; ajmaline; yohimbine; lidocaine; secobarbital; viloxazine; mepivacaine; meperidine; doxylamine; labetalol; temazepam; amodiaquine; benperidol; droperidol; hydroxychloroquine; zolpidem; ketoprofen; alminoprofen; cicletanine; moclobemide; chloroquine; cocaine; timolol; nomifensine; ticlopidine; acenocoumarol; vindesine; mexiletine; dipyridamole; trazodone; pipamperone; pyrimethamine; benazepril; vincristine; metapramine; chlordiazepoxide; oxprenolol; warfarin; clorazepate; flecainide; phencyclidine; thiopental; fenfluramine; metipranolol; triprolidine; naproxen; buprenorphine; verapamil; buspirone; tianeptine; midazolam; bupivacaine; carbinoxamine; loprazolam; cetirizine; chlorpheniramine; moperone; cibenzoline; medifoxamine; astemizole; vinblastine; nicardipine; bisoprolol; diltiazem; glibornuride; reserpine; aconitine; nitrendipine; diazepam; mianserin; ramipril; haloperidol; tetracaine; alprenolol; acepromethazine; glibenclamide; chlorophenacinone; doxepin; nimodipine; diphenhydramine; cyclizine; histapyrrodine; phenylbutazone; demexiptiline; clozapine; proguanil; trifluperidol; medazepam; cyamemazine; bumadizone; suriclone; propranolol; acepromazine; dothiepin; dextromoramide; fenoprofen; dextropropoxyphene; loxapine; betaxolol; propafenone; promethazine; thioproperazine; metha-

done; amoxapine; quinupramine; opipramol; cyproheptadine; brompheniramine; mefenidramine; protriptyline; flurbiprofen; tetrazepam; zorubicin; prazepam; alimemazine; loperamide; imipramine; desipramine; levomepromazine; hydroxyzine; niflumic acid; penbutolol; fluvoxamine; pimozide; daunorubicin; indomethacin; maprotiline; tropatenine; etodolac; fluoxetine; amitriptyline; nortriptyline; tioclomarol; diclofenac; mefloquine; trimipramine; chlorambucil; lidoflazine; ibuprofen; floctafenine; alpidem; loratadine; chlorpromazine; clomipramine; carpipramine; thioridazine; fentiazac; clemastine; mefenamic acid; fluphenazine; prochlorperazine; penfluridol; bepridil; terfenadine; trifluoperazine

REFERENCE

Tracqui,A.; Kintz,P.; Mangin,P. Systematic toxicological analysis using HPLC/DAD, *J.Forensic Sci.*, **1995**, *40*, 254–262.

SAMPLE

Matrix: blood

Sample preparation: 150 μL Plasma + 300 μL 1 mg/mL piroxicam in MeCN, mix, centrifuge at 3500 rpm for 20 min, inject a 10 μL aliquot of the supernatant.

HPLC VARIABLES

Column: 125 × 4 Nucleosil C18
Mobile phase: MeCN:water:acetic acid 58:38:4
Flow rate: 1
Injection volume: 10
Detector: UV 365

CHROMATOGRAM

Internal standard: piroxicam
Limit of detection: 200 ng/mL

KEY WORDS

rat; pharmacokinetics; plasma

REFERENCE

Troconiz,I.F.; Lopez-Bustamente,L.G.; Fos,D. Tenoxicam pharmacokinetics in rats: A population model, *J.Pharm.Sci.*, **1995**, *84*, 1482–1487.

SAMPLE

Matrix: blood, dialysate

Sample preparation: Blood, plasma. 150 μL Blood or plasma + 300 μL 1 mg/mL piroxicam in MeCN, centrifuge at 3500 rpm for 20 min, inject a 10 μL aliquot of the supernatant. Dialysate. 100 μL Dialysate + 200 μL 1 mg/mL piroxicam in MeCN, centrifuge at 3500 rpm for 20 min, inject a 10 μL aliquot of the supernatant.

HPLC VARIABLES

Column: 125 × 4 Nucleosil C18
Mobile phase: MeCN:water:acetic acid 55:38:4
Flow rate: 1
Injection volume: 10
Detector: UV 365

CHROMATOGRAM

Retention time: 1.5
Internal standard: piroxicam (2.2)
Limit of detection: 200 ng/mL

KEY WORDS

rat; whole blood; plasma; pharmacokinetics

REFERENCE

Lopez-Bustamante,L.G.; Troconiz,J.I.; Fos,D. Tenoxicam: acute dose-dependent disposition studies in rats, *J.Pharm.Sci.*, **1993**, *82*, 851–853.

SAMPLE
Matrix: blood, synovial fluid
Sample preparation: Plasma. 500 μL Plasma + 500 μL 500 mM pH 4 phosphate buffer, mix, homogenize, centrifuge at 900 g. Add 1 mL of the mixture to an Extrelut-1 SPE cartridge. Elute with two 5 mL portions of dichloromethane. Evaporate the eluate under a stream of nitrogen at 35°. Dissolve the residue in 120 μL mobile phase by vortexing, inject an aliquot. Plasma, synovial fluid. Condition a 100 mg C18 SPE cartridge (Phenomenex) with 1 mL MeOH, 1 mL water, and 500 μL 500 mM pH 2 phosphate buffer. 500 μL Plasma or synovial fluid + 500 μL 500 mM pH 2 phosphate buffer. Mix, homogenize, centrifuge at 2500 rpm. Add 1 mL of the clean supernatant to the SPE cartridge, wash with 1 mL water, dry with 2 mL air, elute with 1.25 mL MeCN:25% ammonia 90:10. Evaporate under reduced pressure at 35°. Reconstitute the residue in 100 μL MeOH:100 mM pH 8 phosphate buffer 50:50, inject an aliquot.

HPLC VARIABLES
Column: 250 × 4.6 5 μm Hypersil ODS
Mobile phase: MeOH:100 mM pH 6 sodium dihydrogenphosphate buffer 50:50
Flow rate: 1.5
Injection volume: 50-100
Detector: UV 372

CHROMATOGRAM
Retention time: 2.6-2.8
Internal standard: tenoxicam

OTHER SUBSTANCES
Extracted: lornoxicam

KEY WORDS
plasma; mouse; rat; rabbit; dog; monkey; human; SPE; lornoxicam is IS

REFERENCE
Radhofer-Welte,S.; Dittrich,P. Determination of the novel non-steroidal anti-inflammatory drug lornoxicam and its main metabolite in plasma and synovial fluid, *J.Chromatogr.B*, **1998**, *707*, 151–159.

SAMPLE
Matrix: blood, urine
Sample preparation: Add 1 mL whole blood or urine to Toxi-Tube A (Toxi-Lab, Irvine CA), add 3 mL water, mix by gentle inversion for 5 min, centrifuge at 1500 g for 5 min. Remove the organic layer and evaporate it to dryness under a stream of nitrogen at 40°, reconstitute the residue with 50 μL MeCN:water 50:50, vortex for 10 s, centrifuge at 7500 g for 2 min, inject a 10 (urine) or 30 (blood) μL aliquot. (The detector wavelength shown is the wavelength of maximum absorbance. This will not necessarily be the optimal wavelength for the separation. Multiple wavelengths from 200-350 nm can be scanned using a diode-array detector. Otherwise, 220 nm may be a reasonable choice for initial work. Matrix may interfere.)

HPLC VARIABLES
Guard column: 20 mm long Symmetry C18
Column: 250 × 4.6 5 μm Symmetry C8 (Waters)
Mobile phase: Gradient. A was 50 mM pH 3.8 sodium phosphate buffer. B was MeCN. A:B 85:15 for 6.5 min, 65:35 for 18.5 min, 20:80 for 3 min (step gradient), re-equilibrate at initial conditions for 7 min.
Column temperature: 30
Flow rate: 1 for 6.5 min, to 1.5 over 18.5 min, maintain at 1.5 for 3 min (re-equilibrate at 1.5 mL/min)
Injection volume: 10-30
Detector: UV 200.5

CHROMATOGRAM
Retention time: 12.733

KEY WORDS
whole blood

REFERENCE
Gaillard,Y.; Pépin,G. Use of high-performance liquid chromatography with photodiode-array UV detection for the creation of a 600-compound library. Application to forensic toxicology, *J.Chromatogr.A*, **1997**, *763*, 149–163.

SAMPLE
Matrix: solutions

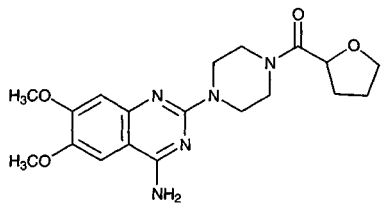
HPLC VARIABLES
Column: 125 × 3 Ecocart LiChrospher 100 RP-18
Mobile phase: Isopropanol:100 mM KH_2PO_4:formic acid 54:100:0.1
Flow rate: 0.6
Detector: UV 254

CHROMATOGRAM
Retention time: 1.5
Limit of quantitation: 200-500 ng/mL

OTHER SUBSTANCES
Simultaneous: acemetacin; diclofenac; flurbiprofen; indomethacin; lonazolac; ketoprofen; naproxen; piroxicam; sulindac

REFERENCE
Baeyens,W.R.G.; Van Der Weken,G.; Van Overbeke,A.; Zhang,Z.D. Preliminary results on the LC-separation of non-steroidal anti-inflammatory agents in conventional and narrow-bore RP set-ups applying columns with different internal diameters, *Biomed.Chromatogr.*, **1995**, *9*, 261–262.

Terazosin



Molecular formula: $C_{19}H_{25}N_5O_4$
Molecular weight: 387.44
CAS Registry No.: 65390-64-7, 70024-40-7 (HCl dihydrate), 63074-08-8 (HCl)
Merck Index: 9297
Lednicer No.: 3 194

SAMPLE
Matrix: blood
Sample preparation: 1 mL Plasma + 100 μL 100 ng/mL IS in water, vortex briefly, add 1 mL 0.9% NaCl and 100 μL 2 M NaOH, vortex briefly. Add 5 mL pentane:dichloromethane 50:50, mix at 40 rpm for 20 min, centrifuge at 500 g for 10 min, freeze the lower aqueous layer in a acetone-dry ice bath, evaporate the organic layer to dryness under a stream of nitrogen. Reconstitute the residue in 100 μL mobile phase, vortex vigorously for 15 s, inject a 65 μL aliquot.

HPLC VARIABLES
Column: 250 × 4.6 10 μm Chiralpak AD (Chiral Technologies, USA)
Mobile phase: Hexane:2-propanol:diethylamine 70:30:0.1
Column temperature: 30
Flow rate: 1
Injection volume: 65
Detector: F em 238 ex 370

CHROMATOGRAM
Internal standard: (+)-glaucine
Limit of quantitation: 500 pg/mL

OTHER SUBSTANCES
Interfering: prazosin

KEY WORDS
plasma; chiral

REFERENCE
Zavitsanos,A.P.; Alebic-Kolbah,T. Enantioselective determination of terazosin in human plasma by normal phase high-performance liquid chromatography--electrospray mass spectrometry, *J.Chromatogr.A*, **1998**, *794*, 45–56.

SAMPLE
Matrix: blood
Sample preparation: 1 mL Plasma + 100 μL 200 ng/mL IS in water, vortex briefly, add 1 mL 0.9% NaCl and 100 μL 2 M NaOH, vortex briefly. Add 5 mL pentane:dichloromethane 50:50, mix at 40 rpm for 20 min, centrifuge at 500 g for 10 min, freeze the lower aqueous layer in a acetone-dry ice bath, evaporate the organic layer to dryness under a stream of nitrogen. Reconstitute the residue in 70 μL hexane:2-propanol 90:10, vortex vigorously for 15 s, inject a 20 μL aliquot.

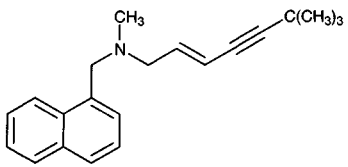
HPLC VARIABLES
Column: 100 × 2.1 10 μm Chiralpak AD (Chiral Technologies, USA)
Mobile phase: Hexane:2-propanol containing 0.05% diethylamine 65:35
Flow rate: 0.15
Injection volume: 20
Detector: MS, HP 1100 electrospray, positive ion mode, drying gas nitrogen 310°, 10 L/min, nebulizer pressure 60 p.s.i.g., quadropole temperature 100°, capillary voltage 4500 V, SIM, m/z 388.2, post-column solvent addition of isopropanol:5 mM ammonium acetate 75:25

CHROMATOGRAM
Retention time: 4.8, 5.8 (enantiomers)
Internal standard: prazosin (4.8)
Limit of quantitation: 62.5 pg/mL

KEY WORDS
plasma; chiral

REFERENCE
Zavitsanos,A.P.; Alebic-Kolbah,T. Enantioselective determination of terazosin in human plasma by normal phase high-performance liquid chromatography--electrospray mass spectrometry, *J.Chromatogr.A*, **1998**, *794*, 45–56.

Terbinafine



Molecular formula: $C_{21}H_{25}N$
Molecular weight: 291.44
CAS Registry No.: 91161-71-6
Merck Index: 9299
Lednicer No.: 4 55

SAMPLE
Matrix: blood, tissue
Sample preparation: Adjust pH of 100 μL plasma or 500 μg tissue to 9.0, add IS, extract with hexane. Extract the organic layer with sulfuric acid:isopropanol, inject an aliquot of this solution.

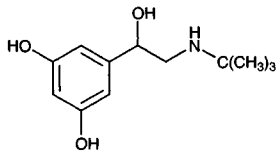
HPLC VARIABLES
Column: 150 × 2 Ultrasphere C18
Mobile phase: MeCN:pH 4.0 phosphate buffer 40:60
Detector: UV 224

CHROMATOGRAM
Limit of detection: 1.1 ng/g (fetal tissue), 2.23 ng/mL (plasma)

KEY WORDS
rabbit; plasma; placenta; fetus

REFERENCE
Gries,W.J.; Wan,W.; Matos,F.J.; de Meireles,J.C.; Pimplaskar,H.K.; Sileno,A.P.; Romeo,V.D.; Xia,W.J.; Behl,C.R. A specific and sensitive method for quantitating buprenorphine hydrochloride in a nasal solution (Abstract 2517), *Pharm.Res.*, **1997**, *14*, S381.

Terbutaline



Molecular formula: $C_{12}H_{19}NO_3$
Molecular weight: 225.29
CAS Registry No.: 23031-25-6, 23031-32-5 (sulfate)
Merck Index: 9302

SAMPLE
Matrix: blood
Sample preparation: Condition a Bond-Elut C18 SPE cartridge with MeCN and water. 1 mL Plasma + 1 mL 20 mM pH 9.0 Na_2HPO_4, mix, add to the SPE cartridge, wash with 5 column volumes of water, dry the SPE cartridge for 5 min, wash with 3 mL dichloromethane:n-butanol 97:3, elute with two 1 mL portions of 0.09% HCl in MeCN, evaporate the eluate to dryness under a stream of nitrogen at 37°, dissolve the residue in 300 μL mobile phase, inject a 200 μL aliquot. (To hydrolyze 3-O-metaproterenol sulfate mix 1 mL plasma and 200 μL 2 μg/mL terbutaline sulfate, add 1 mL 6% trichloroacetic acid, vortex, centrifuge at 1000 g for 15 min. Remove the supernatant and add it to 200 μL 2 M HCl, heat at 65° for 90 min, cool, adjust pH to 10 with 400 μL 2 M carbonate buffer, proceed as above.)

HPLC VARIABLES
Column: 250 × 4.9 5 μm Spherisorb C8
Mobile phase: MeCN:buffer:water 4:1.5:94.5
Flow rate: 1.8
Injection volume: 200
Detector: F ex 200 em 300 (cut-off filter)

CHROMATOGRAM
Retention time: 14.8
Internal standard: terbutaline

OTHER SUBSTANCES
Extracted: metaproterenol

KEY WORDS
SPE; plasma; terbutaline is IS

REFERENCE
Selinger,K.; Hill,H.M.; Matheou,D.; Dehelean,L. Determination of free and total metaproterenol in human plasma by high-performance liquid chromatography with fluorimetric detection, *J.Chromatogr.*, **1989**, *493*, 230–238.

SAMPLE
Matrix: blood
Sample preparation: Inject 1 mL plasma onto column A with mobile phase A, wash with mobile phase A for 1.7 min, backflush contents of column A onto column B with mobile phase B, monitor the effluent from column B.

HPLC VARIABLES
Column: A 10 × 1.5 37 μm Corasil C18; B C18 guard column + 250 × 4.6 10 μm Spherisorb ODS
Mobile phase: A water; B MeOH:67 mM pH 5 phosphate buffer:40 g/L sodium dodecyl sulfate: diethylamine 55:45:0.5:0.02 (before use condition column with 55:45:5:0.05)
Flow rate: 1
Injection volume: 1000
Detector: E, EG & G Princeton Applied Research Model 400 EC, carbon fiber working electrode + 1.3 V, silver phosphate reference electrode (construction details given)

CHROMATOGRAM
Retention time: 7
Limit of detection: 0.8 ng/mL

KEY WORDS
plasma; column-switching

REFERENCE
Sagar,K.A.; Kelly,M.T.; Smyth,M.R. Analysis of terbutaline in human plasma by high-performance liquid chromatography with electrochemical detection using a micro-electrochemical flow cell, *J.Chromatogr.*, **1992**, *577*, 109–116.

SAMPLE
Matrix: blood
Sample preparation: Condition a Bond Elut Si SPE cartridge by washing twice with 1 mL MeOH, twice with 1 mL water, and once with 1 mL 100 mM pH 9.2 K_2HPO_4. Add 1 mL plasma + 100 μL 500 ng/mL atenolol in water, wash twice with 1 mL water, centrifuge at 1000 g for 5 min, elute with 1 mL MeOH. Evaporate MeOH to dryness at 40° under a stream of air and dissolve residue in 200 μL mobile phase, inject an aliquot.

HPLC VARIABLES
Column: 250 × 4.6 Spherisorb S5 SCX
Mobile phase: MeOH:MeCN:water 40:40:20 containing 0.2% perchloric acid (apparent pH 1.7)
Flow rate: 1.5
Injection volume: 100
Detector: F ex 200 no emission filter

CHROMATOGRAM
Retention time: 7
Internal standard: atenolol (13)
Limit of detection: 2500 ng/mL

OTHER SUBSTANCES
Extracted: albuterol
Noninterfering: aminophylline, beclomethasone, cloprednol, dexamethasone, fenoterol, ipratropium bromide, methylprednisolone, orciprenaline, prednisolone, reproterol, rimiterol, salmeterol, sodium cromoglycate, theophylline

KEY WORDS
plasma; SPE

REFERENCE
McCarthy,P.T.; Atwal,S.; Sykes,A.P.; Ayres,J.G. Measurement of terbutaline and salbutamol in plasma by high performance liquid chromatography with fluorescence detection, *Biomed.Chromatogr.*, **1993**, *7*, 25–28.

SAMPLE

Matrix: blood

Sample preparation: 2 mL Whole blood or plasma + 2 mL buffer + 5 mL chloroform:isopropanol: n-heptane 60:14:26, shake gently horizontally for 10 min, centrifuge at 2800 g for 10 min. Remove the lower organic layer and evaporate it to dryness under vacuum at 45°, reconstitute the residue in 100 μL mobile phase, centrifuge at 2800 g for 5 min, inject a 50 μL aliquot of the supernatant. (Buffer was saturated ammonium chloride solution 25% diluted with water, adjusted to pH 9.5 with 25% ammonia solution.)

HPLC VARIABLES

Column: 300 × 3.9 4 μm NovaPack C18

Mobile phase: MeOH:THF:buffer 65:5:30 (Buffer was 0.68 g/L (10 mM (sic)) KH_2PO_4 adjusted to pH 2.6 with concentrated orthophosphoric acid.) (At the end of each session wash the column with water for 1 h and MeOH for 1 h, re-equilibrate for 30 min.)

Column temperature: 30

Flow rate: 0.8

Injection volume: 50

Detector: UV 225

CHROMATOGRAM

Retention time: 3.36

Limit of detection: <120 ng/mL

KEY WORDS

whole blood; plasma; interferences may occur–compounds (all of which are extracted) elute in this order tenoxicam; iproniazid; methocarbamol; methotrexate; caffeine; nialamide; colchicine; cytarabine; benzoylecgonine; acetaminophen; diazoxide; dacarbazine; sulfinpyrazole; flumazenil; sulpride; morphine; atenolol; toloxatone; terbutaline; albuterol; phenobarbital; ranitidine; tiapride; phenol; chlormezanone; aspirin; metformin; ritodrine; codeine; sultopride; amisulpride; naltrexone; lisinopril; benzocaine; nizatidine; nalorphine; mephenesin; naloxone; sotalol; carteolol; procainamide; carbamazepine; bromazepam; nalbuphine; nadolol; procarbazine; dihydralazine; omeprazole; strychnine; acebutolol; glutethimide; chlorpropamide; glipizide; triazolam; prazosin; flunitrazepam; clonazepam; metoclopramide; melphalan; estazolam; tolbutamide; ephedrine; clonidine; pindolol; clobazam; minoxidil; disopyramide; nitrazepam; dextromethorphan; tofisopam; zopiclone; debrisoquine; sulindac; alprazolam; cycloguanil; lorazepam; methaqualone; ketamine; piroxicam; metoprolol; nifedipine; quinine; mephentermine; prilocaine; pentazocine; oxazepam; tiaprofenic acid; quinidine; celiprolol; ajmaline; yohimbine; lidocaine; secobarbital; viloxazine; mepivacaine; meperidine; doxylamine; labetalol; temazepam; amodiaquine; benperidol; droperidol; hydroxychloroquine; zolpidem; ketoprofen; alminoprofen; cicletanine; moclobemide; chloroquine; cocaine; timolol; nomifensine; ticlopidine; acenocoumarol; vindesine; mexiletine; dipyridamole; trazodone; pipamperone; pyrimethamine; benazepril; vincristine; metapramine; chlordiazepoxide; oxprenolol; warfarin; clorazepate; flecainide; phencyclidine; thiopental; fenfluramine; metipranolol; triprolidine; naproxen; buprenorphine; verapamil; buspirone; tianeptine; midazolam; bupivacaine; carbinoxamine; loprazolam; cetirizine; chlorpheniramine; moperone; cibenzoline; medifoxamine; astemizole; vinblastine; nicardipine; bisoprolol; diltiazem; glibornuride; reserpine; aconitine; nitrendipine; diazepam; mianserin; ramipril; haloperidol; tetracaine; alprenolol; acepromethazine; glibenclamide; chlorophenacinone; doxepin; nimodipine; diphenhydramine; cyclizine; histapyrrodine; phenylbutazone; demexiptiline; clozapine; proguanil; trifluperidol; medazepam; cyamemazine; bumadizone; suriclone; propranolol; acepromazine; dothiepin; dextromoramide; fenoprofen; dextropropoxyphene; loxapine; betaxolol; propafenone; promethazine; thioproperazine; methadone; amoxapine; quinupramine; opipramol; cyproheptadine; brompheniramine; mefenidramine; protriptyline; flurbiprofen; tetrazepam; zorubicin; prazepam; alimemazine; loperamide; imipramine; desipramine; levomepromazine; hydroxyzine; niflumic acid; penbutolol; fluvoxamine; pimozide; daunorubicin; indomethacin; maprotiline; tropatenine; etodolac; fluoxetine; amitriptyline; nortriptyline; tioclomarol; diclofenac; mefloquine; trimipramine; chlorambucil; lidoflazine; ibuprofen; floctafenine; alpidem; loratadine; chlorpromazine; clomipramine; carpipramine; thioridazine; fentiazac; clemastine; mefenamic acid; fluphenazine; prochlorperazine; penfluridol; bepridil; terfenadine; trifluoperazine

REFERENCE

Tracqui,A.; Kintz,P.; Mangin,P. Systematic toxicological analysis using HPLC/DAD, *J.Forensic Sci.*, **1995**, *40*, 254–262.

SAMPLE
Matrix: blood, urine
Sample preparation: Add 1 mL whole blood or urine to Toxi-Tube A (Toxi-Lab, Irvine CA), add 3 mL water, mix by gentle inversion for 5 min, centrifuge at 1500 g for 5 min. Remove the organic layer and evaporate it to dryness under a stream of nitrogen at 40°, reconstitute the residue with 50 μL MeCN:water 50:50, vortex for 10 s, centrifuge at 7500 g for 2 min, inject a 10 (urine) or 30 (blood) μL aliquot. (The detector wavelength shown is the wavelength of maximum absorbance. This will not necessarily be the optimal wavelength for the separation. Multiple wavelengths from 200-350 nm can be scanned using a diode-array detector. Otherwise, 220 nm may be a reasonable choice for initial work. Matrix may interfere.)

HPLC VARIABLES
Guard column: 20 mm long Symmetry C18
Column: 250 × 4.6 5 μm Symmetry C8 (Waters)
Mobile phase: Gradient. A was 50 mM pH 3.8 sodium phosphate buffer. B was MeCN. A:B 85:15 for 6.5 min, 65:35 for 18.5 min, 20:80 for 3 min (step gradient), re-equilibrate at initial conditions for 7 min.
Column temperature: 30
Flow rate: 1 for 6.5 min, to 1.5 over 18.5 min, maintain at 1.5 for 3 min (re-equilibrate at 1.5 mL/min)
Injection volume: 10-30
Detector: UV 200.5

CHROMATOGRAM
Retention time: 3.683

KEY WORDS
whole blood

REFERENCE
Gaillard,Y.; Pépin,G. Use of high-performance liquid chromatography with photodiode-array UV detection for the creation of a 600-compound library. Application to forensic toxicology, *J.Chromatogr.A*, **1997**, *763*, 149–163.

SAMPLE
Matrix: formulations
Sample preparation: Tablets, capsules. Mix tablets or capsules with 10 mL water, sonicate 30 min, centrifuge, inject an aliquot. Liquid formulations. Dilute liquid formulations with water, inject an aliquot.

HPLC VARIABLES
Column: 125 × 4 5 μm LiChrospher 100 RP-18 endcapped
Mobile phase: MeOH:water 40:60 containing 2 mM KOH + 10 mM hexanoic acid
Flow rate: 0.4
Injection volume: 20
Detector: UV 214

CHROMATOGRAM
Limit of detection: 1030 ng/mL

OTHER SUBSTANCES
Also analyzed: albuterol, fenoterol

KEY WORDS
tablets; capsules; liquid formulations

REFERENCE
Ackermans,M.T.; Beckers,J.L.; Everaerts,F.M.; Seelen,I.G. Comparison of isotachophoresis, capillary zone electrophoresis and high-performance liquid chromatography for the determination of salbutamol, terbutaline sulphate and fenoterol hydrobromide in pharmaceutical dosage forms, *J.Chromatogr.*, **1992**, *590*, 341–353.

SAMPLE
Matrix: microsomal incubations
Sample preparation: Condition a C18 Sep-Pak SPE cartridge with three 3 mL portions of EtOH, two 3 mL portions of water, and with 3 mL 10 mM pH 7.5 phosphate buffer. Add the incubation mixture to the SPE cartridge, wash with two 3 mL portions of water, elute with two 1 mL portions of EtOH:50 mM pH 8.5 ammonium chloride buffer 95:5. Evaporate the eluate to dryness under a stream of nitrogen at 60°, reconstitute the residue in 500 μL of the initial mobile phase, inject a 200 μL aliquot.

HPLC VARIABLES
Column: 150 × 5 Nucleosil 10SA
Mobile phase: Gradient. A was 250 mM pH 4.6 ammonium acetate buffer. B was MeCN:500 mM pH 4.6 ammonium acetate buffer 50:50. From A:B 90:10 to 10:90 over 20 min.
Flow rate: 1
Injection volume: 200
Detector: UV 254

CHROMATOGRAM
Retention time: 9.5

OTHER SUBSTANCES
Extracted: bambuterol

KEY WORDS
rat; SPE

REFERENCE
Lindberg,C.; Roos,C.; Tunek,A.; Svensson,L.Å. Metabolism of bambuterol in rat liver microsomes: identification of hydroxylated and demethylated products by liquid chromatography mass spectrometry, *Drug Metab.Dispos.*, **1989**, *17*, 311–322.

SAMPLE
Matrix: perfusate, tissue
Sample preparation: Perfusate. 400 μL Lung perfusate + 400 μL 5% perchloric acid, mix, centrifuge, inject a 200 μL aliquot of the supernatant. Tissue. Homogenize lung in 2 volumes of water (Polytron Homogenizer), mix 400 μL homogenate with 400 μL 5% perchloric acid, mix, centrifuge, inject a 200 μL aliquot.

HPLC VARIABLES
Column: 150 × 5 Nucleosil 10SA
Mobile phase: Gradient. A was 19.3 g ammonium acetate and 14.4 mL acetic acid in 1 L water. B was 19.3 g ammonium acetate and 14.4 mL acetic acid in 1 L MeCN:water 50:50. A:B from 90:10 to 10:90 over 20 min, stay at 10:90 for 3 min, return to initial conditions over 3 min, re-equilibrate for 7 min.
Flow rate: 1
Injection volume: 200
Detector: UV 254

CHROMATOGRAM
Retention time: 8

OTHER SUBSTANCES
Extracted: metabolites, bambuterol

KEY WORDS
lung; guinea pig

REFERENCE
Ryrfeldt,Å.; Nilsson,E.; Tunek,A.; Svensson,L.-Å. Bambuterol: uptake and metabolism in guinea pig isolated lungs, *Pharm.Res.*, **1988**, *5*, 151–155.

SAMPLE
Matrix: solutions
Sample preparation: Dilute with 5% dextrose, inject a 40 μL aliquot.

HPLC VARIABLES
Column: Waters microparticulate C18
Mobile phase: MeOH:350 mM acetic acid and 5 mM sodium heptanesulfonate 35:65
Flow rate: 1.6-2.0
Injection volume: 40
Detector: F ex 280 em 310

CHROMATOGRAM
Retention time: 5.33

OTHER SUBSTANCES
Simultaneous: theophylline, methyldopate, isoproterenol

REFERENCE
Williams,D.A.; Fung,E.Y.Y.; Newton,D.W. Ion-pair high-performance liquid chromatography of terbutaline and catecholamines with aminophylline in intravenous solutions, *J.Pharm.Sci.*, **1982**, *71*, 956–958.

SAMPLE
Matrix: solutions
Sample preparation: Prepare a 10 μg/mL solution in MeOH, inject a 20 μL aliquot.

HPLC VARIABLES
Column: 125 × 4.9 Spherisorb S5W silica
Mobile phase: MeOH containing 10 mM ammonium perchlorate and 1 mL/L 100 mM NaOH in MeOH, pH 6.7
Flow rate: 2
Injection volume: 20
Detector: E, LeCarbone, V25 glassy carbon electrode, + 1.2 V

CHROMATOGRAM
Retention time: 1.7

OTHER SUBSTANCES
Also analyzed: acebutolol, acepromazine, acetophenazine, N-acetylprocainamide, albuterol, alprenolol, amethocaine, amiodarone, amitriptyline, antazoline, atenolol, azacyclonal, bamethan, benactyzine, benperidol, benzethidine, benzocaine, benzoctamine, benzphetamine, benzquinamide, bromhexine, bromodiphenhydramine, bromperidol, brompheniramine, brompromazine, buclizine, bufotenine, bupivacaine, buprenorphine, butacaine, butethamate, chlorcyclizine, chlorpheniramine, chlorphenoxamine, chlorprenaline, chlorpromazine, chlorprothixene, cimetidine, cinchonidine, cinnarizine, clemastine, clomipramine, clonidine, cocaine, cyclazocine, cyclizine, cyclopentamine, cyproheptadine, deserpidine, desipramine, dextromoramide, dextropropoxyphene, dicyclomine, diethylcarbamazine, diethylpropion, diethylthiambutene, dihydroergotamine, dimethindene, dimethothiazine, diphenhydramine, diphenoxylate, dipipanone, diprenorphine, dipyridamole, disopyramide, dothiepin, doxapram, doxepin, doxylamine, droperidol, ephedrine, ergocornine, ergocristine, ergocristinine, ergocryptine, ergometrine, ergosine, ergosinine, ergotamine, ethopropazine, etorphine, etoxeridine, fenethazine, fenfluramine, fenoterol, fentanyl, flavoxate, fluopromazine, flupenthixol, fluphenazine, flurazepam, haloperidol, hydroxyzine, hyoscine, ibogaine, imipramine, indapamine, iprindole, isothipendyl, isoxsuprine, ketanserin, laudanosine, lidocaine, lofepramine, loxapine, maprotiline, mecamylamine, meclophenoxate, meclozine, medazepam, mephentermine, mepivacaine, meptazinol, mepyramine, mesoridazine, metaraminol, methadone, methamphetamine, methapyrilene, methdilazene, methotrimeprazine, methoxamine, methoxyphenamine, methoxypromazine, methylephedrine, methylergonovine, methysergide, metoclopramide, metopimazine, metoprolol, mianserin, morazone, nadolol, nalorphine, naloxone, naphazoline, nicotine, nifedipine, nomifensine, nortriptyline, noscapine, orphenadrine, oxeladin, oxprenolol, oxymetazolin, papaverine, pargyline, pecazine, penbutolol, pentazocine, penthienate, pericyazine, perphenazine, phenadoxone, phenampromide, phenazocine, phenbutrazate, phendimetrazine, phenelzine, phenglutarimide, phenindamine, pheniramine, phenmetrazine, phenomorphan, phenoperidine,

phenothiazine, phenoxybenzamine, phentolamine, phenylephrine, phenyltoloxamine, physostigmine, piminodine, pimozide, pindolol, pipamazine, pipazethate, piperacetazine, piperidolate, pipradol, pirenzepine, piritramide, pizotifen, practolol, pramoxine, prazosin, prenylamine, prilocaine, primaquine, proadifen, procainamide, procaine, prochlorperazine, procyclidine, proheptazine, prolintane, promazine, promethazine, pronethalol, properidine, propiomazine, propranolol, prothipendyl, protriptyline, proxymetacaine, pseudoephedrine, pyrimethamine, quinidine, quinine, ranitidine, rescinnamine, sotalol, tacrine, terazosin, terfenadine, thenyldiamine, theophylline, thiethylperazine, thiopropazate, thioproperazine, thioridazine, thiothixene, thonzylamine, timolol, tocainide, tolpropamine, tolycaine, tranylcypromine, trazodone, trifluoperazine, trifluperidol, trimeperidine, trimeprazine, trimethobenzamide, trimethoprim, trimipramine, tripelennamine, triprolidine, tryptamine, verapamil, xylometazoline

REFERENCE

Jane,I.; McKinnon,A.; Flanagan,R.J. High-performance liquid chromatographic analysis of basic drugs on silica columns using non-aqueous ionic eluents. II. Application of UV, fluorescence and electrochemical oxidation detection, *J.Chromatogr.*, **1985**, *323*, 191–225.

SAMPLE

Matrix: solutions

HPLC VARIABLES

Column: 250 × 4.6 Chirex 3020 (Phenomenex)
Mobile phase: Hexane:1,2-dichloroethane:EtOH/trifluoroacetic acid 55:35:10 (EtOH/trifluoroacetic acid was premixed 20:1.)
Flow rate: 0.7-1
Injection volume: 20
Detector: UV 278

KEY WORDS

chiral; α = 1.28 for enantiomers

REFERENCE

Cleveland,T. Pirkle-concept chiral stationary phases for the HPLC separation of pharmaceutical racemates, *J.Liq.Chromatogr.*, **1995**, *18*, 649–671.

SAMPLE

Matrix: solutions
Sample preparation: Inject an aliquot of a 200 μM solution in MeOH.

HPLC VARIABLES

Column: 100 × 4.7 7 μm Hypercarb (Shandon)
Mobile phase: MeOH containing 5 mM N-benzyloxycarbonylglycyl-L-proline and 4.5 mM NaOH
Column temperature: 17
Injection volume: 20
Detector: UV 270

CHROMATOGRAM

Retention time: k' 3.6 (first enantiomer)

KEY WORDS

chiral; α = 1.17

REFERENCE

Huynh,N.-H.; Karlsson,A.; Pettersson,C. Enantiomeric separation of basic drugs using N-benzyloxycarbonylglyclyl-L-proline as counter ion in methanol, *J.Chromatogr.A*, **1995**, *705*, 275–287.

SAMPLE

Matrix: solutions

HPLC VARIABLES

Column: 250 × 4.6 5 μm Supelcosil LC-DP (A) or 250 × 4 5 μm LiChrospher 100 RP-8 (B)

Mobile phase: MeCN:0.025% phosphoric acid:buffer 25:10:5 (A) or 60:25:15 (B) (Buffer was 9 mL concentrated phosphoric acid and 10 mL triethylamine in 900 mL water, adjust pH to 3.4 with dilute phosphoric acid, make up to 1 L.)
Flow rate: 0.6
Injection volume: 25
Detector: UV 229

CHROMATOGRAM
Retention time: 5.62 (A), 3.27 (B)

OTHER SUBSTANCES
Also analyzed: acebutolol, acepromazine, acetaminophen, acetazolamide, acetophenazine, albuterol, alprazolam, amitriptyline, amobarbital, amoxapine, antipyrine, atenolol, atropine, azatadine, baclofen, benzocaine, bromocriptine, brompheniramine, brotizolam, bupivacaine, buspirone, butabarbital, butalbital, caffeine, carbamazepine, cetirizine, chlorcyclizine, chlordiazepoxide, chlormezanone, chloroquine, chlorpheniramine, chlorpromazine, chlorpropamide, chlorprothixene, chlorthalidone, chlorzoxazone, cimetidine, cisapride, clomipramine, clonazepam, clonidine, clozapine, cocaine, codeine, colchicine, cyclizine, cyclobenzaprine, dantrolene, desipramine, diazepam, diclofenac, diflunisal, diltiazem, diphenhydramine, diphenidol, diphenoxylate, dipyridamole, disopyramide, dobutamine, doxapram, doxepin, droperidol, encainide, ethidium bromide, ethopropazine, fenoprofen, fentanyl, flavoxate, fluoxetine, fluphenazine, flurazepam, flurbiprofen, fluvoxamine, furosemide, glutethimide, glyburide, guaifenesin, haloperidol, homatropine, hydralazine, hydrochlorothiazide, hydrocodone, hydromorphone, hydroxychloroquine, hydroxyzine, ibuprofen, imipramine, indomethacin, ketoconazole, ketoprofen, ketorolac, labetalol, levorphanol, lidocaine, loratadine, lorazepam, lovastatin, loxapine, mazindol, mefenamic acid, meperidine, mephenytoin, mepivacaine, mesoridazine, metaproterenol, metformin, methadone, methdilazine, methocarbamol, methotrexate, methotrimeprazine, methoxamine, methyldopa, methylphenidate, metoclopramide, metolazone, metoprolol, metronidazole, midazolam, moclobemide, morphine, nadolol, nalbuphine, naloxone, naphazoline, naproxen, nifedipine, nizatidine, norepinephrine, nortriptyline, oxazepam, oxycodone, oxymetazoline, paroxetine, pemoline, pentazocine, pentobarbital, pentoxifylline, perphenazine, pheniramine, phenobarbital, phenol, phenolphthalein, phentolamine, phenylbutazone, phenyltoloxamine, phenytoin, pimozide, pindolol, piroxicam, pramoxine, prazepam, prazosin, probenecid, procainamide, procaine, prochlorperazine, procyclidine, promazine, promethazine, propafenone, propantheline, propiomazine, propofol, propranolol, protriptyline, quazepam, quinidine, quinine, racemethorphan, ranitidine, remoxipride, risperidone, salicylic acid, scopolamine, secobarbital, sertraline, sotalol, spironolactone, sulfinpyrazone, sulindac, temazepam, terfenadine, tetracaine, theophylline, thiethylperazine, thiopental, thioridazine, thiothixene, timolol, tocainide, tolbutamide, tolmetin, trazodone, triamterene, triazolam, trifluoperazine, triflupromazine, trimeprazine, trimethoprim, trimipramine, verapamil, warfarin, xylometazoline, yohimbine, zopiclone

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KEY WORDS
details of plasma extraction

REFERENCE
Koves,E.M. Use of high-performance liquid chromatography-diode array detection in forensic toxicology, *J.Chromatogr.A*, **1995**, *692*, 103–119.

SAMPLE
Matrix: solutions
Sample preparation: Inject a 20 μL aliquot of an 8 μg/mL solution.

HPLC VARIABLES
Column: 250 × 4 4 μm Superspher 100 RP-18
Mobile phase: Buffer containing 10 mM β-cyclodextrin substituted with 2-hydroxy-3-trimethylammoniumpropyl groups (Roquette Frères, Lestrem, France) (Buffer was 0.8% triethylamine adjusted to pH 5.9 with acetic acid.)
Column temperature: 22.5
Flow rate: 0.8
Injection volume: 20
Detector: UV 275

CHROMATOGRAM
Retention time: 18.02, 19.88 (enantiomers)

KEY WORDS
chiral

REFERENCE
Roussel,C.; Favrou,A. Cationic β-cyclodextrin: a new versatile chiral additive for separation of drug enantiomers by high-performance liquid chromatography, *J.Chromatogr.A*, **1995**, *704*, 67–74.

SAMPLE
Matrix: solutions
Sample preparation: Prepare a 1-10 μg/mL solution in water, inject an aliquot.

HPLC VARIABLES
Column: 250 × 4.6 5 μm Hypersil SCX/C18
Mobile phase: MeCN:25 mM pH 3 Na_2HPO_4 50:50
Injection volume: 20
Detector: UV 254

CHROMATOGRAM
Retention time: k' 3.39

OTHER SUBSTANCES
Also analyzed: amitriptyline, barbital, benzoic acid, butabarbital, clomipramine, clonazepam, desipramine, diazepam, flurazepam, furosemide, imipramine, nitrazepam, phenobarbital, phenol, phenolphthalein, pindolol, propranolol, resorcinol, salicylic acid, secobarbital, xylazine

KEY WORDS
effect of mobile phase pH on capacity factor is discussed

REFERENCE
Walshe,M.; Kelly,M.T.; Smyth,M.R.; Ritchie,H. Retention studies on mixed-mode columns in high-performance liquid chromatography, *J.Chromatogr.A*, **1995**, *708*, 31–40.

SAMPLE
Matrix: solutions

HPLC VARIABLES
Column: 150 × 4.6 Zorbax phenyl
Mobile phase: MeCN:water 15:85 containing 10 mM KH_2PO_4, adjusted to pH 3.1 with phosphoric acid
Flow rate: 1.5
Detector: UV 212

CHROMATOGRAM
Retention time: 2.2

REFERENCE
Liu,P.; Bergstrom,T.K. Quantitative evaluation of aqueous isopropyl alcohol enhancement on skin flux of terbutaline (sulfate). 2. Permeability contributions of equilibrated drug species across human skin in vitro, *J.Pharm.Sci.*, **1996**, *85*, 320–325.

SAMPLE
Matrix: tissue
Sample preparation: 300 mg Skin + propranolol, homogenize with 5 mL MeOH four times, combine the homogenates, filter. Evaporate the filtrate to dryness, reconstitute in mobile phase, inject a 5 μL aliquot.

HPLC VARIABLES
Column: 250 × 4.6 5 μm Spherisorb cyano
Mobile phase: MeCN:pH 5.6 buffer 30:70
Flow rate: 1.4
Injection volume: 5
Detector: UV 225

CHROMATOGRAM
Retention time: 3.3
Internal standard: propranolol hydrochloride (7.9)
Limit of detection: 100 ng/mL

KEY WORDS
stability-indicating; skin

REFERENCE
Tenjarla,S.N.; Allen,R.; Mitchell,B. High-performance liquid chromatographic assay of terbutaline for preformulation studies, *J.Liq.Chromatogr.*, **1995**, *18*, 1603–1615.

SAMPLE
Matrix: urine
Sample preparation: Condition a Bond Elut C18 SPE cartridge with 3 volumes of MeOH and 2 volumes of water, dry under vacuum. Add 500 μL urine to the SPE cartridge, wash with 5 volumes of water, elute with 200 μL MeOH:50 mM pH 6 potassium phosphate buffer 50:50, add 50 μL 50 mM Na_3PO_4 to the eluate, pass argon through the mixture, inject a 25 μL aliquot.

HPLC VARIABLES
Column: 300 mm long μBondapak phenyl
Mobile phase: MeCN:50 mM pH 5 phosphate buffer 6:94
Flow rate: 2.8
Injection volume: 25
Detector: F ex 280 em 310

CHROMATOGRAM
Retention time: 4.1
Internal standard: terbutaline

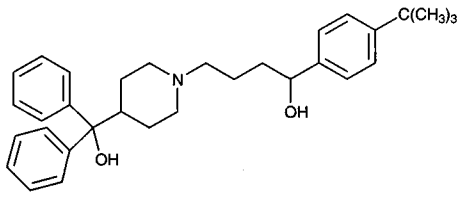
OTHER SUBSTANCES
Extracted: metaproterenol

KEY WORDS
SPE; protect from light; terbutaline is IS

REFERENCE
MacGregor,T.R.; Nastasi,L.; Farina,P.R.; Keirns,J.J. Isolation and characterization of metaproterenol-3-O-sulfate: a conjugate of metaproterenol in human urine, *Drug Metab.Dispos.*, **1983**, *11*, 568–573.

Terfenadine

Molecular formula: $C_{32}H_{41}NO_2$
Molecular weight: 471.68
CAS Registry No.: 50679-08-8
Merck Index: 9307
Lednicer No.: 4 48, 104



SAMPLE
Matrix: blood